

Three new species of comb-tailed spiders (Araneae: Hahniidae) from a Mexican oak forest with comments on their natural history and sexual behavior

M. Antonio Galán-Sánchez¹ and Fernando Alvarez-Padilla^{1,2}: ¹Laboratorio de Aracnología, Facultad de Ciencias, Universidad Nacional Autónoma de México s/n Ciudad Universitaria, México D. F. Del. Coyoacán, Código postal 04510, México; ²Research Associate, Department of Entomology, California Academy of Sciences, San Francisco, California, USA. E-mail: fap@ciencias.unam.mx

Abstract. Three new hahniid species, *Hahnia quadriseta* sp. nov., *Neoantistea multidentata* sp. nov. and *N. aspeubira* sp. nov., are described from a total of 1,131 individuals collected during spider inventories in Veraeruz, Mexico. All specimens were collected inside four plots of *Quercus* forest, from leaf litter using active searching, sifted litter processed with Berlese funnels and pitfall traps. Sexual behavior and the natural history of *N. aspeubira* sp. nov. are analyzed. In addition, the stridulatory organ morphology of both *Neoantistea* species is described. Stridulatory organs are absent in *Hahnia quadriseta* sp. nov., although present in other species of this genus. More information regarding this inventory and additional views of specimens can be found online at <http://www.unamfcaracnolab.com>

Keywords: Taxonomy, RTA clade, Nearctic, Mexico, stridulatory organ.

ZooBank publication: <http://zoobank.org/?lsid=urn:lsid:zoobank.org:pub:C2D143C3-BA0A-4CF8-8CFF-E4117529328D>

The family Hahniidae includes small, entelegyne ecribellate spiders, some of which are characterized by a distinctive transverse arrangement of the spinnerets. Hahniidae have a wide distribution and comprise 249 species included in 28 genera, with a high diversity in the Holarctic and Paleotropical regions (World Spider Catalog 2017). The most important taxonomic synopses of this group were made by Opell & Beatty (1976) for all Nearctic species and Forster (1970) for the New Zealand fauna; since then, no revisions have been made for these or any other regions, including Mexico. Recent, smaller taxonomic studies on this family have focused on the Palaearctic and Paleotropical faunas, describing a considerable number of species for *Hahnia* C. L. Koch, 1841, *Alistra* Thorell, 1894 and the new genus *Pacifantistea* Marusik, 2011 from eastern Russia (Chen et al. 2003; Zhang & Zhang 2003; Chen et al. 2009; Marusik 2011; Zhang et al. 2011, 2013; Zhang & Zhang 2013; Liu et al. 2015; Suguro 2015).

The taxonomy and systematics of the family Hahniidae have proven to be a challenge that remains unresolved. All phylogenetic studies place Hahniidae inside the RTA clade (Coddington & Levi 1991; Wheeler et al. 2016); however, its rank as a family (Bertkau 1878) has been debated throughout its taxonomic history. Some authors have considered it a subfamily of Agelenidae (Emerton 1890; Simon 1892; Bristowe 1938; Gertsch 1949; Bonnet 1959; Kaestner 1968); while other researchers considered it a separate family (Petrunkevitch 1933; Gerhardt & Kaestner 1938; Kaston 1948).

The position of Hahniidae at more inclusive taxonomic levels has also presented several challenges. Based upon the number of cardiac ostia and extension of trachea into the cephalothorax, this family was first considered a member of the Lycosoidea (Gerhardt & Kaestner 1938; Kaston 1948). Later, Hahniidae was included inside Amaurobioidea based upon the male pedipalp anatomy and the presence of unbranched trachea (Lehtinen 1967). Finally, this family was included in Dictynoidea (Forster 1970; Coddington & Levi

1991) based upon a detailed analysis of the tracheal system morphology (Forster 1970). However, the inclusion of Hahniidae within these superfamilies, although based in each instance on a thorough examination of their anatomy, only considered separate character systems in isolation, without phylogenetic analysis.

Molecular phylogenetic analyses (Spagna & Gillespie 2008; Miller et al. 2010; Spagna et al. 2010) questioned Forster's Dictynoidea. A recent, comprehensive, multi-gene analysis (Wheeler et al. 2016) has radically rearranged the RTA clade, including Forster's Dictynoidea, Cryphoecinae, formerly Hahniidae, have been transferred to Cybaeidae, leaving only Hahniinae and Cybaeolinae confirmed as hahniids. Hahniidae appear related to a relimited Dictynidae and a resurrected Toxopidae, and these, in turn, to an enlarged Cybaeidae (Wheeler et al. 2016, fig. 5).

Currently Hahniidae (*sensu* Wheeler et al. 2016) includes two subfamilies: Hahniinae and Cybaeolinae, which can be differentiated by the arrangement of their spinnerets. Of these subfamilies, Hahniinae is well represented in the Nearctic region with 27 species and a distribution extending from Canada to southwest Mexico. Twelve species are known to inhabit the Mexican territory. Seven species are described from both sexes, four only by females and one by two male specimens. The distribution of these species within Mexico and information indicating if they are represented by one or both sexes is presented in Table 1.

Web construction, sexual behavior and other aspects of the biology in hahniids have been understudied. For Nearctic Hahniidae, Opell & Beatty (1976) reviewed the natural history of *Neoantistea* Gertsch, 1934 species and described their microhabitats, web-building behavior, egg sac morphology and phenology. Likewise, morphological analysis of stridulatory structures and their implications for behavior have been poorly analyzed. Stridulatory organs have been reported for several genera such as *Antistea* Simon, 1898, *Neoantistea*, *Hahnia* and *Pacifantistea*; however, most of the studies only

Table 1.—Diversity of Hahniidae in Mexico. Abbreviations: B. C. N. = Baja California Norte.

Genus	Species	Distribution	Description		
			♂ ♀	♀	♂
<i>Neoantistea</i>	<i>N. pueblensis</i> Opell & Beatty, 1976	Puebla	X		
	<i>N. mulaiki</i> Gertsch, 1946	Tamaulipas	X		
	<i>N. lyrica</i> Opell & Beatty, 1976	Hidalgo	X		
	<i>N. inaffecta</i> Opell & Beatty, 1976	Colima	X		
	<i>N. jacalana</i> Gertsch, 1946	Hidalgo		X	
	<i>N. hidalgoensis</i> Opell & Beatty, 1976	Hidalgo		X	
	<i>N. spica</i> Opell & Beatty, 1976	Distrito Federal		X	
	<i>N. inifistula</i> Opell & Beatty, 1976	Veracruz		X	
<i>Hahnia</i>	<i>H. cinerea</i> Emerton, 1890	Puebla	X		
	<i>H. sanjuanensis</i> Exline, 1938	B. C. N.	X		
	<i>H. nobilis</i> Opell & Beatty, 1976	Hidalgo	X		
	<i>H. veracruzana</i> Gertsch & Davis, 1940	Veracruz			X

mentioned the presence of these organs (Harm 1966; Forster 1970; Brignoli 1976, 1977, 1978; Opell & Beatty 1976; Benoit 1978; Bosmans & Thijs 1980; Marusik 2011; Zhang et al. 2011, 2013; Zhang & Zhang, 2013). Only one study documented the stridulatory organs with scanning electron microscope (SEM) images (Joequé & Bosmans 1982). The latter paper described and discussed the differences between these organs in *H. eburneensis* Joequé & Bosmans, 1982 and 16 other species of *Hahnia*, identifying five stridulatory organ types on the chelicerae retrolateral surface, concluding that the presence of several types of stridulatory organs in this genus could offer characters to separate closely related species. It is important to mention that *H. quadriseta* sp. nov. did not present any kind of stridulatory structure.

The three species described here were collected as part of a spider inventory done in Mexican *Quercus* forests, and are described from a total of 1,131 individuals. In addition, the natural history, sexual behavior and stridulatory organs of *N. aspebira* sp. nov. and the detailed anatomy of the stridulatory organ of *N. multidentata* sp. nov. are described and discussed.

METHODS

Sampling and spider collection.—The specimens were collected as part of a spider inventory following standardized protocols (Coddington et al. 1991) inside two *Quercus* oak forests: one near the boundary of the Pico de Orizaba National Park, ca. two km southwest from Atotonilco de Calcahualco, Veracruz, Mexico, from May 2012 to February 2013; and another at Xamatiepac de Calcahualco, ca. 23 km east from Pico de Orizaba National Park, Veracruz, Mexico, from April 2013 to February 2014. All specimens were collected inside four plots of one hectare each. Atotonilco: Plot I with central coordinates 19°8'17.4" N, 97°12'16.2" W and an elevation of 2,300 m; Plot II 19°8'30.2" N, 97°12'21.5" W and 2,388 m. Xamatiepac: Plot I 19°7'34.1" N, 97°4'1.5" W and 1,710 m; Plot II 19°7'32.5" N, 97°4'3.2" W and 1,700 m. All species were collected from leaf litter with active searching, and from sifted leaf litter processed with Berlese funnels and pitfall traps.

Morphological methods and material examined.—All specimens were collected and stored in 96% ethanol. Photographs of external anatomy were taken with a Nikon DS-Fi2 camera connected to a Nikon SMZ1000 dissecting microscope and a Nikon E200 Microscope under LED illumination. Multi-focal images were captured using Nikon (NIS Elements 4.0) software and montage processing done with Helicon Focus (5.3.14). Female genitalia were dissected and digested following the protocol of Alvarez-Padilla & Hormiga (2007 (2008)), cleared with clove oil and mounted in temporary preparations (Coddington 1983). Cleared genitalia were analyzed and photographed using the Nikon E200 Microscope. Illustrations were done with Nikon Y-IDT drawing tube. SEM samples were coated with gold and images taken with a JEOL JSM-5310LV SEM. All measurements were done using a micrometric ocular graticule and are given in millimeters. Left male pedipalps were illustrated and photographed. Leg measurements are shown as: total length (femur, patella and tibia, metatarsus, tarsus).

Additional information regarding these inventory methods and high-resolution versions of the images are available online at <http://www.unamfcaracnolab.com> (Alvarez-Padilla Laboratory 2014). Type material is deposited in the Colección Nacional de Arácnidos (CNAN), Instituto de Biología, Universidad Nacional Autónoma de México. Additional examined material is deposited in the Laboratorio de Aracnología Collection, Facultad de Ciencias, (CAFC-UNAM).

Behavioral observations.—Live spiders were collected from the study plots and kept individually inside plastic cages (width x length x height: 5 × 8 × 5 cm; 6 × 9 × 6 cm) with leaf litter from the same place. The spiders were fed with *Drosophila melanogaster* (nubbin strain) once a week. The description of behavior was based on the direct observation of 10 specimens (2 ♂, 7 ♀, 1 immature) and took place from 27 October to 30 November 2015. All sexual behaviors were observed at room temperature that averaged 19.8 ± 1.8°C. The courtship was observed during dark conditions (01:32 AM) under red light.

The following abbreviations are used in the text and figures: ALE, anterior lateral eyes; ALS, anterior lateral

spinnerets; AME, anterior median eyes; B, bursae; Bg, Bennett's glands; Bh, basal hematodochae; Cd, copulatory ducts; Co, copulatory openings; E, embolus; Eb, embolus base; Fd, fertilization ducts; LS, lower sheet; ML, middle layer; Pa, patellar apophysis; PLE, posterior lateral eyes; PLS, posterior lateral spinnerets; PME, posterior median eyes; PMS, posterior median spinnerets; O, refuge opening; RTA, retrolateral tibial apophysis; RTM, retrolateral tibial macroseta; S, spermathecae; SEM, scanning electron microscope; Sd, spermatid ducts; Ss, secondary spermathecae; T, tegulum; US, upper sheet.

TAXONOMY

Family *Hahniidae* Bertkau, 1878

Subfamily *Hahniinae* Bertkau, 1878

Genus *Hahnia* C. L. Koch, 1841

Hahnia quadriseta sp. nov.

<http://zoobank.org/?lsid=urn:lsid:zoobank.org:act:D3E256B0-F181-4894-8B16-11D8E738C991>

Figs. 1–17, 55

Type material.—*Holotype male*. MEXICO: *Veracruz*: Atonileo de Calcahualco, ca. 15 km from Pico de Orizaba Volcano, Plot II, 19°8'30.2" N, 97°12'21.5" W, elev. 2,388 m, oak forest, leaf litter, collected with pitfall traps, 4–14 October 2012 (CNAN-T1138).

Paratypes. MEXICO: *Veracruz*: 1 ♀ allotype, same data as holotype (CNAN-T1139); 2 ♂, 1 ♀, same data except 21–30 May 2012, pitfall trap (CAFC-UNAM); 1 ♀, same locality data except Plot I, 19°8'17.4" N, 97°12'16.2" W, elev. 2,300 m, 21–30 May 2012, F.A. Rivera-Quiroz (CACF-UNAM); 3 ♂, same data except 4–14 October 2012, Berlese funnel (CNAN).

Other material examined.—MEXICO: *Veracruz*: 1 ♂, Atonileo de Calcahualco, 15 km from Pico de Orizaba Volcano, Plot I, 19°8'17.4" N, 97°12'16.2" W 2,300 m, 21–30 May 2012, U. Garcilazo-Cruz (CAFC-UNAM); 2 ♂, 1 ♀, same data except 4–14 October 2012, U. Garcilazo-Cruz (CAFC-UNAM); 2 ♂, 3 ♀, same data except 15–24 February 2013, F. Alvarez-Padilla (CAFC-UNAM); 2 ♂, 5 ♀, same locality data except Plot II, 19°8'30.2" N, 97°12'21.5" W, 2,388 m, 21–30 May 2012, Berlese funnel (CAFC-UNAM); 1 ♂, 1 ♀, same data except 4–14 October 2012, pitfall trap (CAFC-UNAM); 2 ♂, 3 ♀, same data except 15–24 February 2013, Berlese funnel (CAFC-UNAM); 1 ♀, Xamactipac de Calcahualco, Plot I, 19°7'34.1" N, 97°4'1.5" W, 1,710 m, 19–27 April 2013, F. J. Salgueiro-Sepúlveda (CAFC-UNAM); 2 ♂, 1 ♀, same data except 2–11 October 2013, D. Piedra-Jimenez (CAFC-UNAM); 1 ♂, same data except 4–17 February 2014, M. Servín-Pastor (CAFC-UNAM); 2 ♀, same locality data except Plot II, 19°7'32.5" N, 97°4'3.2" W, 1,700 m, 4–17 February 2014, Berlese funnel (CAFC-UNAM).

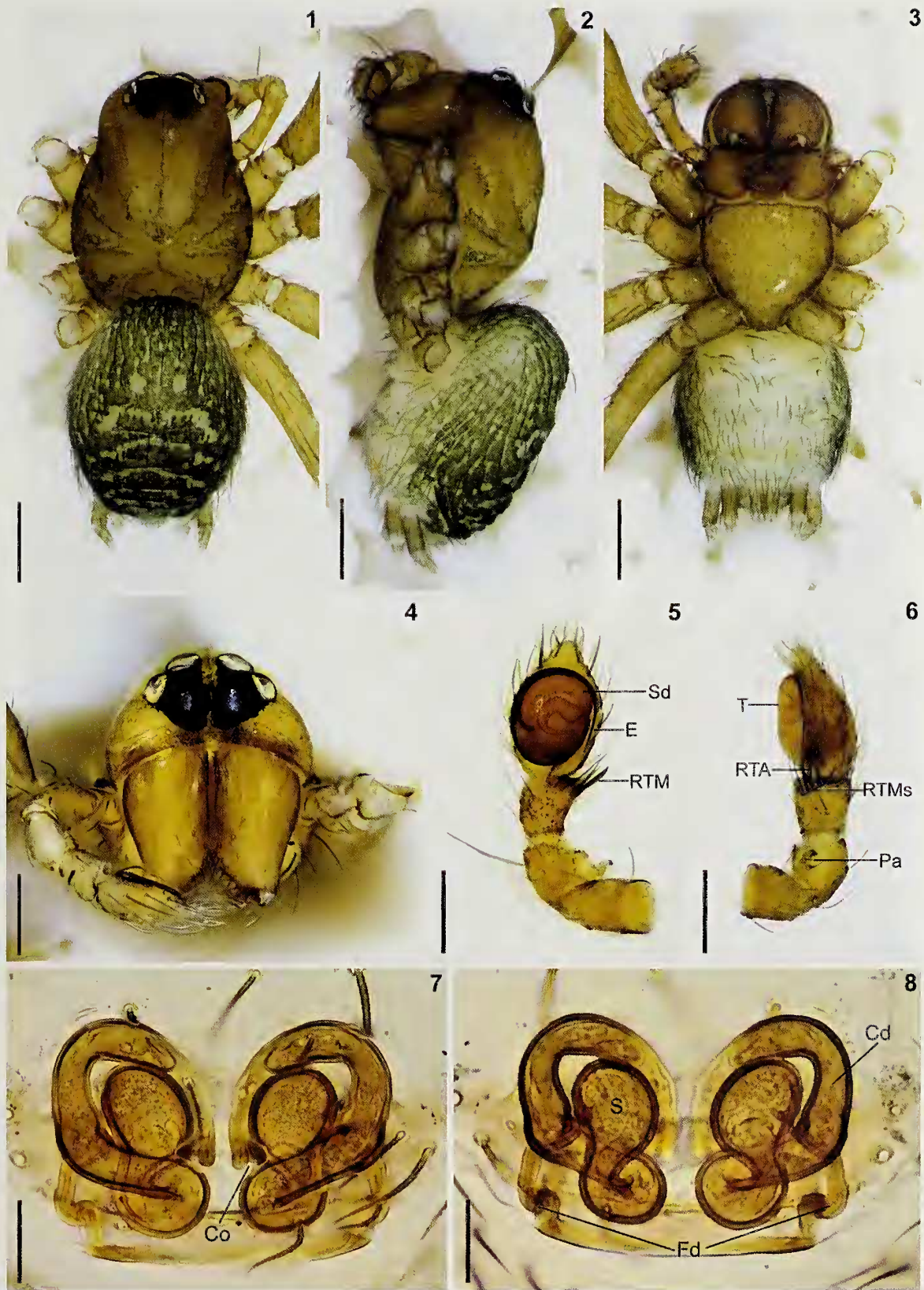
Etymology.—The specific epithet is a noun in apposition from the Latin *quadrus* (four) and *seta* (hair or bristle), referring to the presence of four macrosetae on the retrolateral surface of the male palpal tibia.

Diagnosis.—*Hahnia quadriseta* sp. nov. is similar to *H. senaria* Zhang, Li & Zheng, 2011 in having only six eyes and by having the same somatic appearance (Figs. 1, 4; see also Zhang et al. 2011: figs. 22, 23). Both sexes differ from *H.*

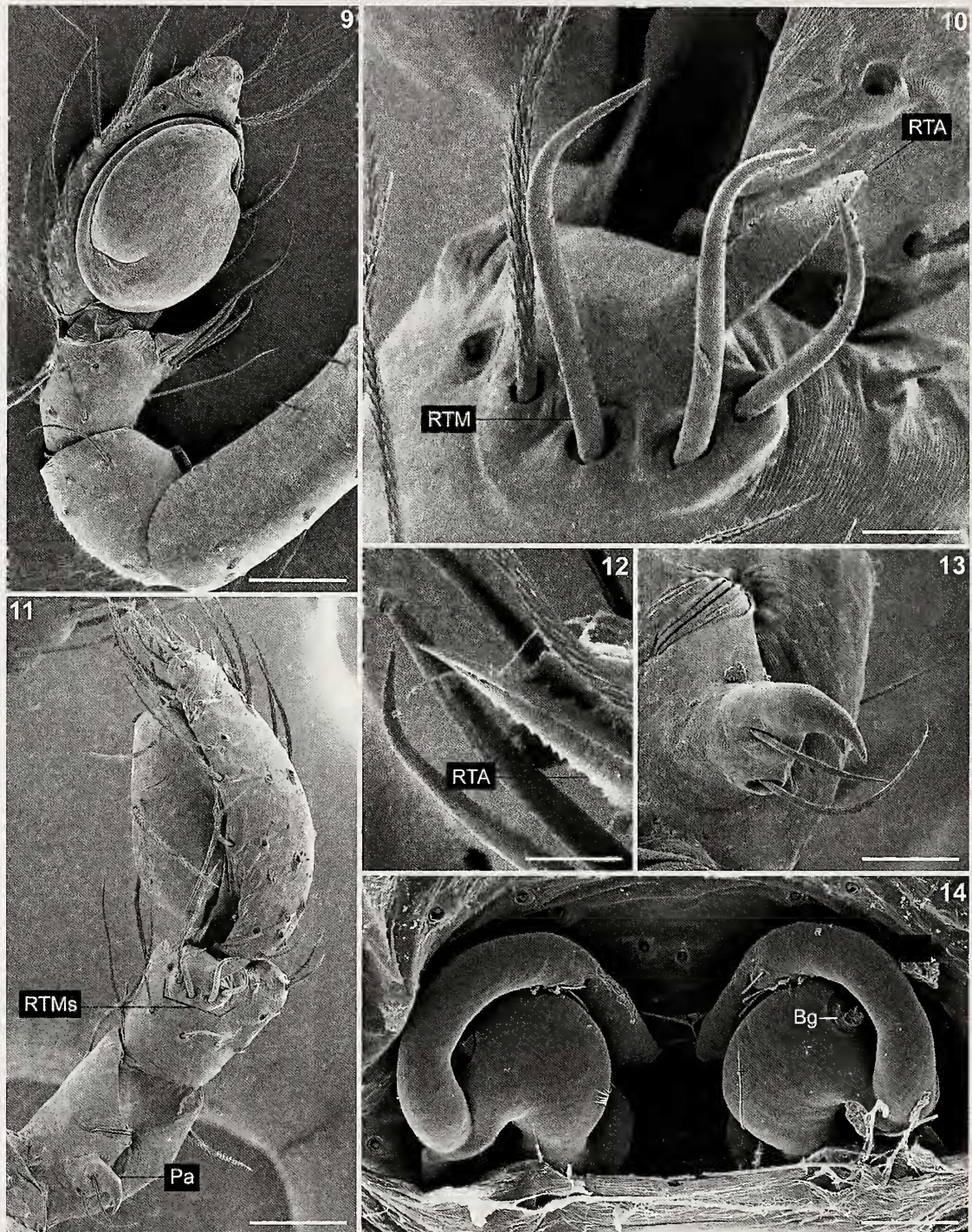
senaria by the position of the tracheal spiracle closer to the base of the median spinnerets (Fig. 3). Females of this species differ by the presence of two conspicuous copulatory openings located medially, and by the long copulatory ducts and long fertilization ducts originating laterally on the spermathecae (Figs. 7, 8, 16, 17). Males of this species differ by the presence of four macrosetae slightly curved apically on the retrolateral surface of the palpal tibia (RTMs) (Figs. 10, 11), and by the short and slightly serrated RTA (Figs. 10, 12).

Description.—*Male* (Holotype). Total length 1.4. Cephalothorax 0.57 long, 0.42 wide. Carapace dorsal surface light brown, slightly darker in pars cephalica; pars thoracica with a reticulated dark pattern concentrated behind the pars cephalica and over the radial furrows (Fig. 1); pear-shaped, narrowed anteriorly, highest point of carapace located between ocular region and fovea. Fovea indistinct; stridulatory organs absent. Ocular region 0.12 long, 0.20 wide, dark color, with setae located between PLE-PME and PME-PME. Six eyes with an arrangement of two triads, posterior eye row procurved in frontal view (Fig. 4). Eye sizes and interdistances: ALE, PLE and PME equal in size (0.05); ALE-ALE equal to PME-PME (0.03), ALE-PLE equal to PLE-PME (0.01). Clypeus less than a diameter of ALE, 0.04. Chelicerae 0.22 long, 0.12 wide, yellow-brown, retrolateral edges slightly concave apically, two promarginal and two retromarginal teeth. Endites 0.13 long, 0.14 wide, same color and pattern as the chelicerae, convergent apically, retrolateral edges concave, serrula darker in coloration. Labium 0.05 long, 0.09 wide, same color pattern as the endites, with strong basal notches. Sternum 0.30 long, 0.30 wide, same color pattern as carapace, margin darker. Shield-shaped, truncated anteriorly, narrowed posteriorly, margins undulated and slightly elevated between coxae (Fig. 3). Abdomen oval, covered with setae, raised anteriorly above posterior region of carapace (Fig. 2), stridulatory organs absent. Dorsal pattern greyish, with five chevron-like markings that extend towards the posterior edge and two semicircular spots located in anterior region (Fig. 1). Ventral surface white. Colulus indistinct. Spinnerets light brown; PMS 0.07, ALS proximal segment 0.09, distal segment 0.03, PLS proximal segment 0.10, distal segment 0.08. Legs: I 1.33 (0.40) (0.45) (0.25) (0.23); II 1.19 (0.36) (0.39) (0.24) (0.20); III 1.09 (0.31) (0.34) (0.24) (0.20); IV 1.36 (0.40) (0.44) (0.28) (0.24); leg formula: 4123; light brown; all femora with ventral row of five to six macrosetae; patella and tibia I to IV dorsal surface with macroseta; tarsi without macrosetae. Pedipalp: Patella larger than tibia. Patella with a short retro-basal apophysis, wider at base, bifurcated and hook-shaped (Figs. 11, 13, 15). Tibia wider anteriorly. Cymbium oval, longer than wide, tip slightly elongated. Embolus originating pro-basally, extending around the tegulum and ending near at the base of cymbium (Fig. 15). Tegulum disk-shaped, spermatid duct following the tegular margin and with three turns before entering the embolus (Figs. 5, 15).

Female (Allotype). Total length 1.3. As in male except as noted: Cephalothorax: Carapace 0.48 long, 0.37 wide. Ocular region 0.10 long, 0.15 wide. Clypeus low 0.03. Endites 0.11 long, 0.13 wide. Sternum 0.27 long, 0.27 wide. Abdomen: Tracheal system with a broad spiracle, closer to spinnerets than to epigastric furrow, internally composed of two short



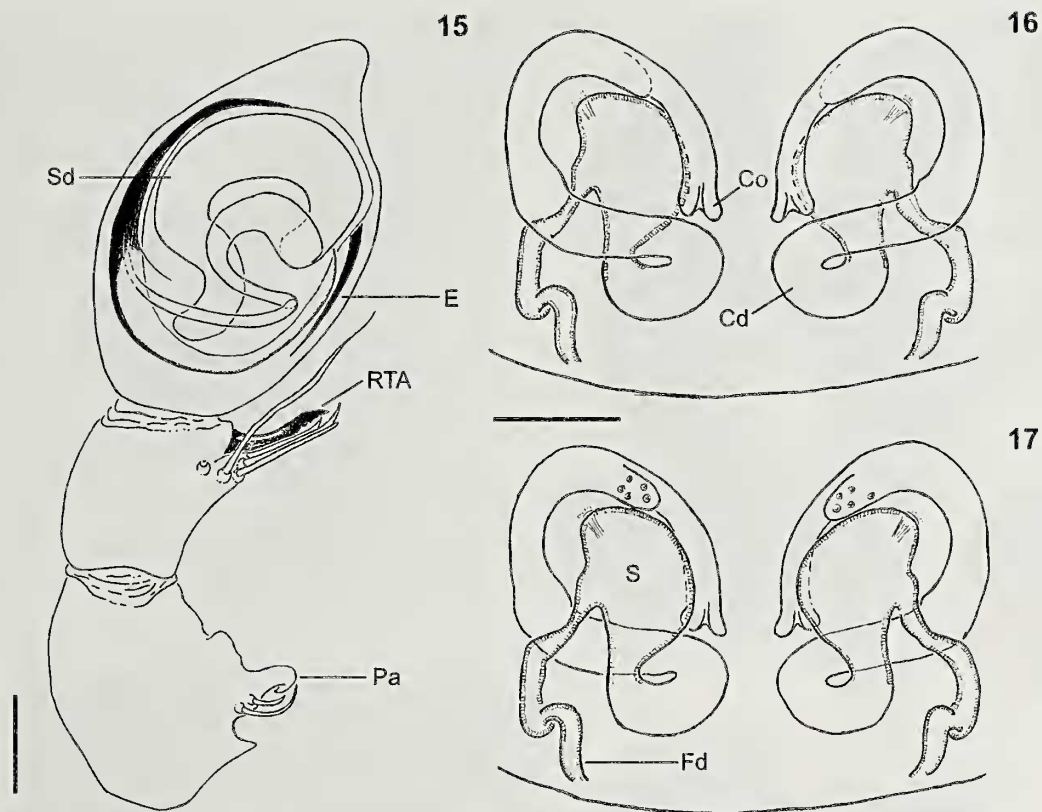
Figures 1–8.—*Hahnia quadriseta* sp. nov.: 1–6, male; 7–8, female. 1, Habitus dorsal view; 2, same lateral view; 3, same ventral view; 4, prosoma anterior view; 5, left pedipalp ventral view; 6, same retrolateral view; 7, epigynum ventral view (cleared); 8, epigynum dorsal view (cleared). Scale bars = 0.5 mm except cleared epigyna = 0.05 mm. Cd, copulatory duct; Co, copulatory opening; E, embolus; Fd, fertilization duct; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; RTMs, retrolateral tibial macrosetae; Sd, spermatheca; S, spermathecae; T, tegulum.



Figures 9–14.—*Hahnia quadriseta* sp. nov.: 9–13, male; 14, female. 9, Pedipalp ventral view; 10, retrolateral surface of pedipalp; 11, pedipalp retrolateral view; 12, RTA dorsal view; 13, patellar apophysis retrolateral view; 14, epigynum dorsal view. Scale bars = 0.01 mm. Bg, Bennett's glands; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; RTMs, retrolateral tibial macrosetae.

tubular tracheae branching in tracheoles that extend towards the cephalothorax (Fig. 55). Distal segment of PLS 0.06. Legs: I 1.10 (0.35) (0.36) (0.20) (0.19); II 0.99 (0.30) (0.32) (0.19) (0.18); III 0.93 (0.29) (0.29) (0.19) (0.16); IV 1.16 (0.35) (0.38) (0.24) (0.19). Epigynum flat, copulatory ducts long, approximately half as wide as spermathecae and surrounding them

completely and forming a posterior coil (Figs. 7, 16). Spermathecae oval, fertilization ducts nearly straight, longer than spermathecae and less than half the spermathecae width and crossing over the copulatory ducts in dorsal view (Figs. 8, 17). The Bennett's glands are located in the anterior region of the spermathecae (Fig. 14). The presence of the glandular



Figures 15–17.—*Hahnia quadriseta* sp. nov., illustrations: 15, male left pedipalp ventral view; 16, cleared epigynum ventral view; 17, cleared epigynum dorsal view. Scale bars = 0.05 mm. Cd, copulatory duct; Co, copulatory opening; E, embolus; Fd, fertilization duct; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; Sd, spermatheca; S, spermathecae.

pores and ducts forming a “lump” over the copulatory ducts could indicate that this region is the secondary spermathecae (Figs. 14, 17).

Variation ($n = 30$).—Males total length 1.2–1.4. Females total length 1.2–1.5. Copulatory openings vary in position from the medial region to the anterior region. Spermathecae vary in orientation from vertical to slightly inclined relative to the epigastric furrow. Specimen coloration varies in its pigmentation from light brown to dark brown. Five specimens presented a dorsal surface of the abdomen without chevron-like markings. Several specimens presented damaged eyes due to physical trauma.

Distribution.—Oak forest in the Atotonilco and Xamatícac de Calchualco regions, Veracruz, Mexico (Figs. 72–74).

Biology.—This species was the most abundant of the family Hahniidae in our inventory. Male and female individuals were collected all year long; however, females were more abundant from May to February, while males were more abundant in February. The presence of a larger number of adult specimens in February could indicate that the breeding season is during winter, gradually decreasing towards spring and again increasing approaching autumn. Juveniles are mainly found in early spring and late summer. Most specimens were caught by cryptic searching (610 individuals), followed by sifting leaf litter processed with Berlese funnels (298 individuals) and pitfall traps (58 individuals). Ten

specimens were found upon vegetation either by direct collecting or beating.

Genus *Neoantistea* Gertsch, 1934

Neoantistea multidentata sp. nov.

<http://zoobank.org/?lsid=urn:lsid:zoobank.org:act:3555CCA7-5301-463D-BAAA-DB9010A80162>

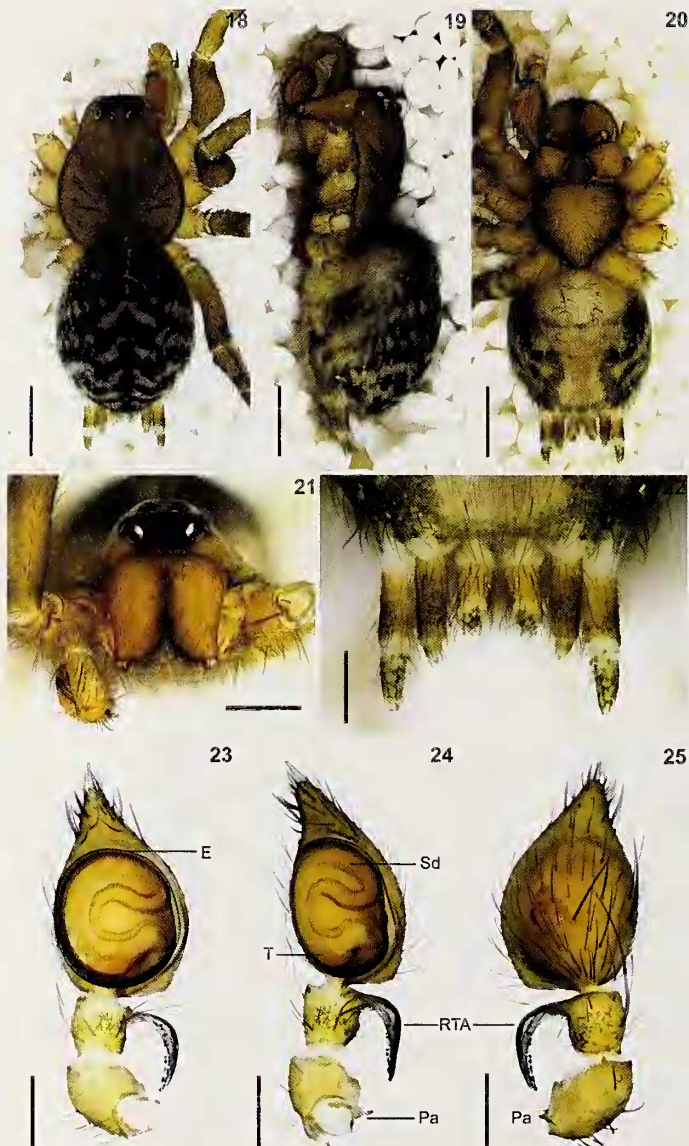
org:act:3555CCA7-5301-463D-BAAA-DB9010A80162

Figs. 18–32, 50, 51, 56, 63, 67–70

Type material.—*Holotype male*. MEXICO: Veracruz: Atotonilco de Calchualco, ca. 15 km from the Pico de Orizaba Volcano, Plot I, 19°8'17.4" N, 97°12'16.2" W, elev. 2,300 m, oak forest, leaf litter, collected with cryptic searching, 4–14 October 2012, F.M. Labarque (CNAN-T1140).

Paratypes. MEXICO: Veracruz: 1 ♀ allotype, same data as holotype, U.G. Cruz (CNAN-T1141); 1 ♂, 1 ♀, Atotonilco de Calchualco, Plot I, 19°8'17.4" N, 97°12'16.2" W, elev. 2,300 m, 4–14 October 2012, M.A. Hernández-Patricio (CAFC-UNAM); 1 ♂, 1 ♀, same data except 15–24 February 2013, F.M. Labarque (CAFC-UNAM).

Other material examined.—MEXICO: Veracruz: 3 ♂, 10 ♀, Atotonilco de Calchualco, 15 km away from the Pico de Orizaba Volcano, Plot I, 19°8'17.4" N, 97°12'16.2" W, 2,300 m, 4–14 October 2012, U. Garcilazo-Cruz, T. Silva, D. Polotow, F.M. Labarque (CAFC-UNAM); 1 ♂, 1 ♀, same



Figures 18–25.—*Neantistea multidentata* sp. nov., male: 18, habitus dorsal view; 19, same lateral view; 20, same ventral view; 21, prosoma anterior view; 22, spinnerets ventral view; 23, left pedipalp ventral view; 24, same retrolateral view; 25, same dorsal view. Scale bars = 0.2 mm except habitus = 0.5 mm. E, embolus; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; Sd, spermatheca; T, tegulum.

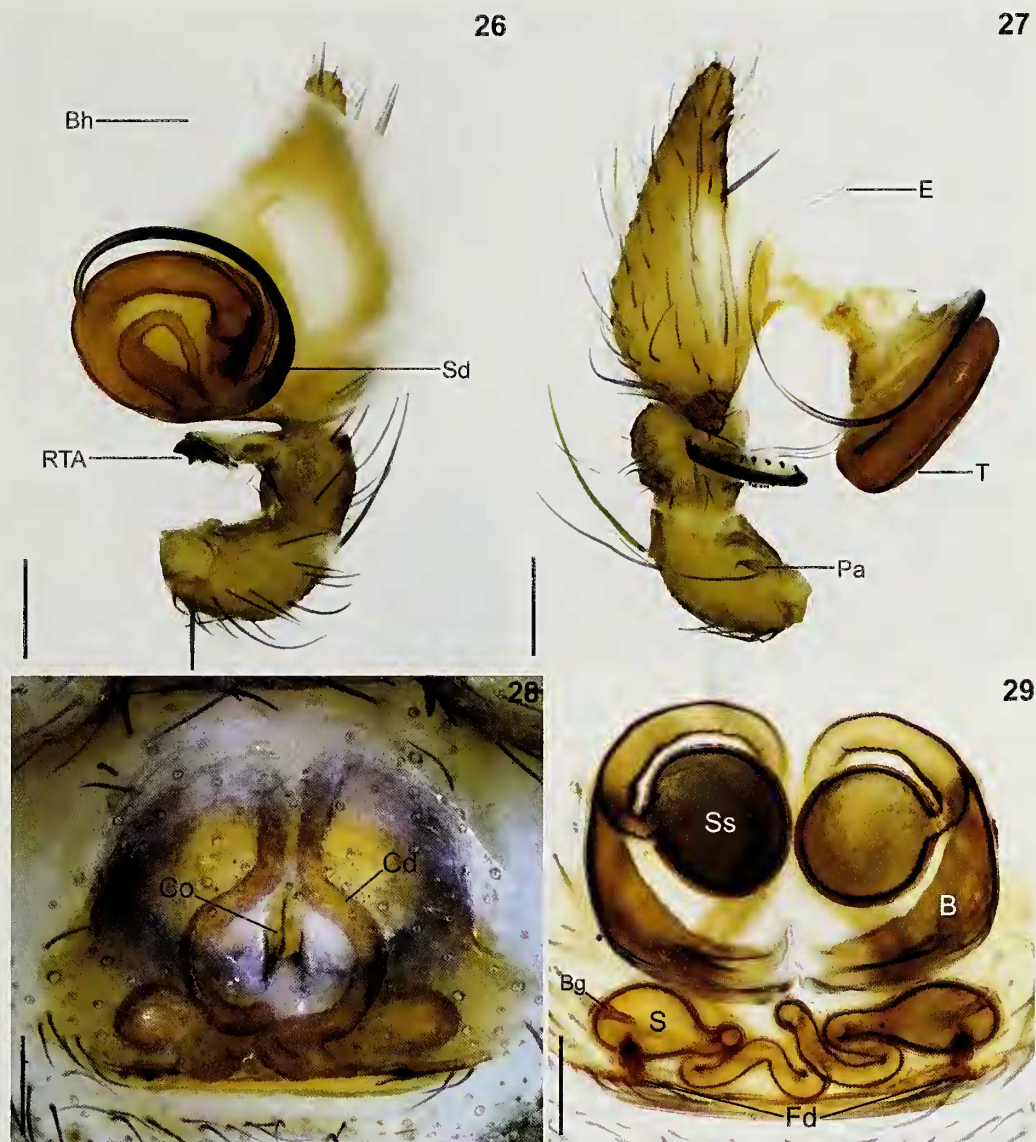
data except 15–24 February 2013, Berlese funnel, U. Garcilazo-Cruz (CAFC-UNAM).

Etymology.—The specific epithet is a combination of adjectives from the Latin *multi* (many) and *dentatus* (toothed), referring to the presence of lines of numerous teeth on the RTA.

Diagnosis.—*Neantistea multidentata* sp. nov. is similar to *N. riparia* (Keyserling, 1887), *N. procteri* Gertsch, 1946, *N. coconino* Chamberlin & Ivie, 1942, *N. gosiuta* Gertsch, 1934, *N. mulaiki* Gertsch, 1946, *N. crandalli* Gertsch, 1946 and *N. santana* Chamberlin & Ivie, 1942 in having the RTA strongly curved proximally (Figs. 24, 26; Opell & Beatty 1976: figs. 24,

26, 30, 40, 46, 56, 62), but differs from these species by having two discontinuous lines of teeth on the inner margins of the RTA (Figs. 25, 30), a long and filiform embolus originating retro-basally, and the distal segment of the PLS shorter than the proximal segment (Figs. 22, 23, 27). The epigynum of *N. multidentata* sp. nov. has a unique anatomy characterized by wide and strongly sclerotized bursae, originating laterally from the secondary spermathecae and oriented towards the medial region (Fig. 29), and semicircular secondary spermathecae, almost as wide as the bursae and located near the anterior margin of the epigynum (Fig. 29).

Description.—*Male* (Holotype). Total length 2.8. Cephalothorax 1.6 long, 1.1 wide. Carapace dorsal surface brown; pars thoracica with a reticulated dark pattern concentrated behind the pars cephalica and over the radial furrows (Fig. 18); pear-shaped, narrowed anteriorly, fovea longitudinal and brown; inconspicuous stridulatory organ on posterior margin composed of rough area with tiny flattened hooks (Fig. 63). Ocular region 0.20 long, 0.37 wide, slightly dark. Eight eyes, both eye rows slightly procurved in frontal view (Fig. 21). Eye sizes and interdistances: AME, PLE and PME equal in size (0.08), ALE (0.1); AME-AME, AME-ALE, ALE-PLE equal in distance (0.01), PLE-PME (0.03), PME-PME (0.06). Clypeus greater than a diameter of AME, 0.12. Chelicerae 0.48 long, 0.24 wide, reddish brown; retrolateral edges slightly concave apically, three promarginal and four retromarginal teeth. Endites 0.26 long, 0.30 wide, light brown, convergent apically, slightly elongated posteriorly, serrula darker in coloration. Labium 0.13 long, 0.20 wide, same color pattern as the carapace, with strong basal notches. Sternum 0.56 long, 0.62 wide, light brown in medial region, margin darker, shield-shaped, truncated anteriorly, narrowed posteriorly, lateral margins with narrow lobes opposite coxae (Fig. 20). Abdomen oval, covered with setae, raised anteriorly above posterior region of carapace, with stridulatory organ dorsolateral to the pedicel composed of two groups of numerous slender and flat macrosetae (Fig. 65). Dorsal pattern greyish-black, with five chevron-like markings that extend towards the postero-lateral region and two reddish semicircular spots located in anterior region (Fig. 18). Ventral surface white. Colulus indistinct. Spinnerets light brown, ALS and PLS dark distally; PMS (0.15), ALS proximal segment (0.15), distal segment (0.04), PLS proximal segment (0.20), distal segment (0.10). Legs: I 3.05 (0.85) (0.95) (0.75) (0.50); II 2.99 (0.88) (0.99) (0.64) (0.48); III 2.72 (0.75) (0.83) (0.66) (0.48); IV 3.51 (0.91) (1.30) (0.80) (0.50); leg formula: 4123; dark brown, all femora, tibiae and metatarsi with proximal and distal dark rings; patella and tibia I to IV dorsal surface with macroseta; tarsi without dark rings or macrosetae. Pedipalp: Patella larger than tibia. Patella with retro-basal apophysis, wider at its base, tapering distally and curved apically (Fig. 51). Retro-basal apophysis flanked by two macrosetae; one smaller than this apophysis, the other considerably longer (Fig. 51). RTA strongly curved, tip bent dorsally and five straight setae at its base (Figs. 25, 50). Cymbium oval, longer than wide, tip elongated. Embolus originating retro-basally and extending around the tegulum making a full turn (Fig. 30). Tegulum disk-shaped, spermatheca following the tegular margin and with three turns before entering the embolus (Figs. 23, 26, 30).

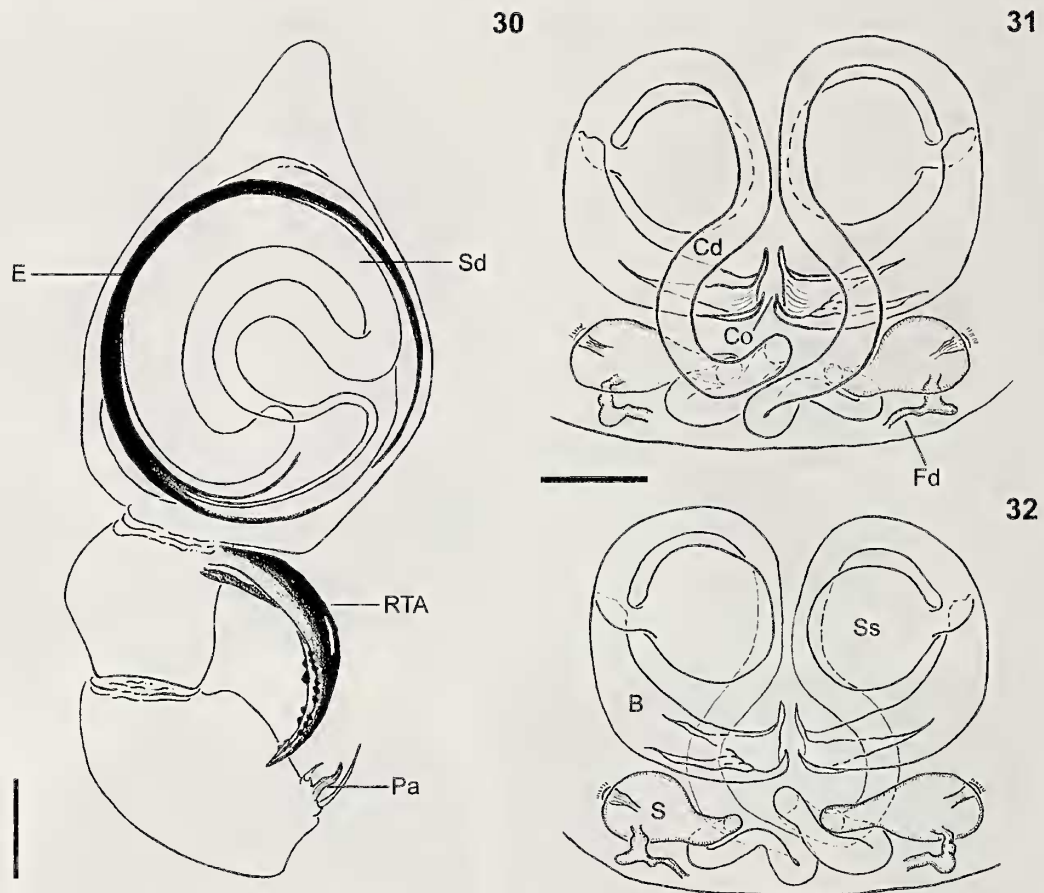


Figures 26–29.—*Neoantistea multidentata* sp. nov. 26, Expanded right male pedipalp, ventral view; 27, same retrolateral view; 28, female epigynum ventral view; 29, same dorsal view (cleared). Scale bars = 0.2 mm (Figs. 26–28), 0.1 mm (Fig. 29). B, bursae; Bg, Bennett's glands; Bh, basal hematodochae; Cd, copulatory duct; Co, copulatory opening; Fd, fertilization duct; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; Sd, spermatic duct; S, spermathecae; Ss, secondary spermathecae; T, tegulum.

Female (Allotype). Total length 3.4. As in male except as noted: Cephalothorax: Carapace 2.0 long, 1.3 wide. Ocular region 0.20 long, 0.38 wide. Eyes and interdistances: ALE (0.09), AME-AME (0.02), PME-PME (0.07). Chelicerae 0.59 long, 0.28 wide. Endites 0.30 long, 0.28 wide. Sternum 0.60 long, 0.70 wide. Abdomen: Tracheal system with distinctive spiracle closer to epigastric furrow than to spinnerets, internally composed of four short tubular tracheae branching in tracheoles that extend towards the posterior and anterior region of the body (Fig. 56). PMS 0.20, ALS proximal segment 0.20, distal segment 0.03. Legs: I 3.13 (0.75) (1.3) (0.60) (0.48); II 2.88 (0.80) (1.0) (0.63) (0.45); III 2.79 (0.83) (0.90) (0.63) (0.43); IV 3.6 (1.0) (1.2) (0.85) (0.55). Epigynum flat, copulatory ducts opening inside a central atrium located between the primary and secondary spermathecae (Fig. 28).

Copulatory ducts visible in ventral view, path surrounding the secondary spermathecae and with several coils before entering the primary spermathecae (Figs. 28, 29). Primary spermathecae oval, located posterior to the secondary spermathecae and smaller than these (Fig. 29). Fertilization ducts short and thick, originating posteriorly from the primary spermathecae. The Bennett's glands are located in the lateral region of the primary spermathecae. (Figs. 29, 32).

Variation ($n = 15$).—Males total length 2.5–2.8. Females total length 3.1–3.4. Coils of ducts vary considerably in orientation. Spermathecae vary in orientation from horizontal to slightly inclined relative to the epigastric furrow. Specimen coloration varies in pigmentation from reddish brown to dark-brown. One specimen presented seven eyes.



Figures 30–32.—*Neoantistea multidentata* sp. nov., illustrations: 30, male left pedipalp ventral view; 31, cleared female epigynum ventral view; 32, same dorsal view. Scale bars = 0.1 mm. B, bursae; Cd, copulatory duct; Co, copulatory opening; E, embolus; Fd, fertilization duct; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; Sd, spermatic duct; S, spermathecae; Ss, secondary spermathecae.

Distribution.—Oak forest in the Atotonilco de Calchahualco region, Veracruz, Mexico (Figs. 72–74).

Biology.—Male and female individuals were collected in October (11 females and four males) and February (only two individuals collected). The presence of adult specimens in October could indicate that the breeding season is during autumn, declining rapidly toward winter (where the time of egg laying and hatching could occur) and rising again toward the autumn. It is likely that juvenile populations are found in early spring and late summer because adult specimens are not found during May and there is a remarkably low number of adults in February. Most specimens were caught in leaf litter with cryptic searching (16 individuals) and only one specimen was found by Berlese funnel.

Neoantistea aspembira sp. nov.

<http://zoobank.org/?lsid=urn:lsid:zoobank.org:act:43E98FC4-99A7-4483-B45B-876917321272>

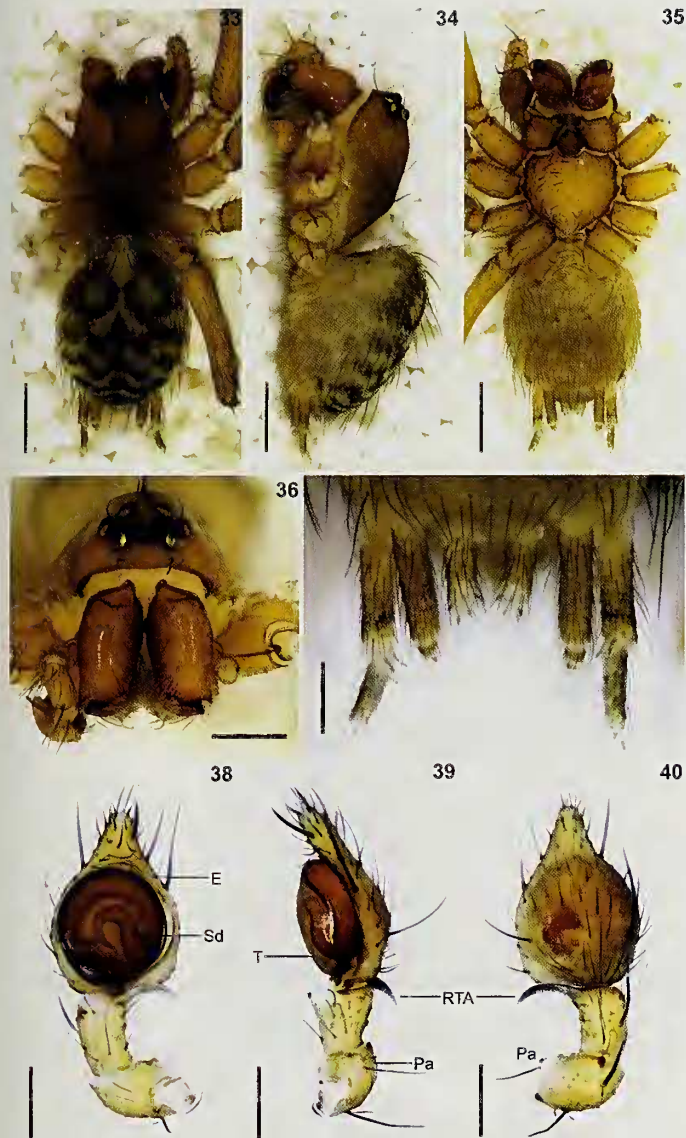
org:act:43E98FC4-99A7-4483-B45B-876917321272

Figs. 33–49, 52–54, 57–62, 64, 66, 71

Type material.—*Holotype male*. MEXICO: Veracruz: Atotonilco de Calchahualco, ca. 15 km from the Pico de Orizaba Volcano, Plot I, 19°8'17.4" N, 97°12'16.2" W, elev. 2,300 m, oak forest, leaf litter, collected with pitfall traps, 21–30 May 2012 (CNAN-T1142).

Paratypes. MEXICO: Veracruz: 1 ♀ allotype, same data as holotype except Plot II, 19°8'30.2" N, 97°12'21.5" W, elev. 2,388 m (CNAN-T1143); 1 ♀, same data as holotype except F. Alvarez-Padilla (CAFC-UNAM); 1 ♂, same data except 4–14 October 2012 (CAFC-UNAM); 1 ♀, same data except Plot II, 19°8'30.2" N, 97°12'21.5" W, elev. 2,388 m, 21–30 May 2012, F.A. Rivera-Quiroz (CAFC-UNAM); 1 ♂, same data except 15–24 February 2013 (CAFC-UNAM).

Other material examined.—MEXICO: Veracruz: 2 ♀, Atotonilco de Calchahualco, 15 km away from the Pico de Orizaba Volcano, Plot I, 19°8'17.4" N, 97°12'16.2" W, 2,300 m, 21–30 May 2012, pitfall trap (CAFC-UNAM); 2 ♂, same data except 4–14 October 2012 (CAFC-UNAM); 1 ♂, 1 ♀, same data except 15–24 February 2013, F.A. Rivera-Quiroz (CAFC-UNAM); 2 ♂, 3 ♀, same data except Plot II, 19°8'30.2" N, 97°12'21.5" W, 2,388 m, 21–30 May 2012, pitfall trap (CAFC-UNAM); 3 ♀, same data except 4–14 October 2012, T. Silva (CAFC-UNAM); 1 ♂, 1 ♀, same data except 15–24 February 2013, U. Garcilazo-Cruz (CAFC-UNAM); 1 ♂, Xamaetipac de Calchahualco, Plot I, 19°7'34.1" N, 97°4'1.5" W, 1,710 m, 19–27 April 2013, pitfall trap (CAFC-UNAM); 1 ♂, same data except 2–11 October 2013, F.A. Rivera-Quiroz (CAFC-UNAM); 1 ♂, same data except 4–17 February 2014, pitfall trap, (CAFC-UNAM); 1 ♂, Plot



Figures 33–40.—*Neoantistea aspembira* sp. nov., male: 35, habitus dorsal view; 36, same lateral view; 37, same ventral view; 38, prosoma anterior view; 39, spinnerets ventral view; 40, left pedipalp ventral view; 41, same retrolateral view; 42, same dorsal view. Scale bars = 0.2 mm except habitus = 1 mm. E, embolus; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; Sd, spermatid duct; T, tegulum.

II, 19°7'32.5" N, 97°4'3.2" W, 1,700 m, 19–27 April 2013, Berlese funnel (CAFC-UNAM); 1 ♂, 2 ♀, same data except 2–11 October 2013, U. Garcilazo-Cruz, (CAFC-UNAM); 1 ♀, same data except 4–17 February 2014 (CAFC-UNAM).

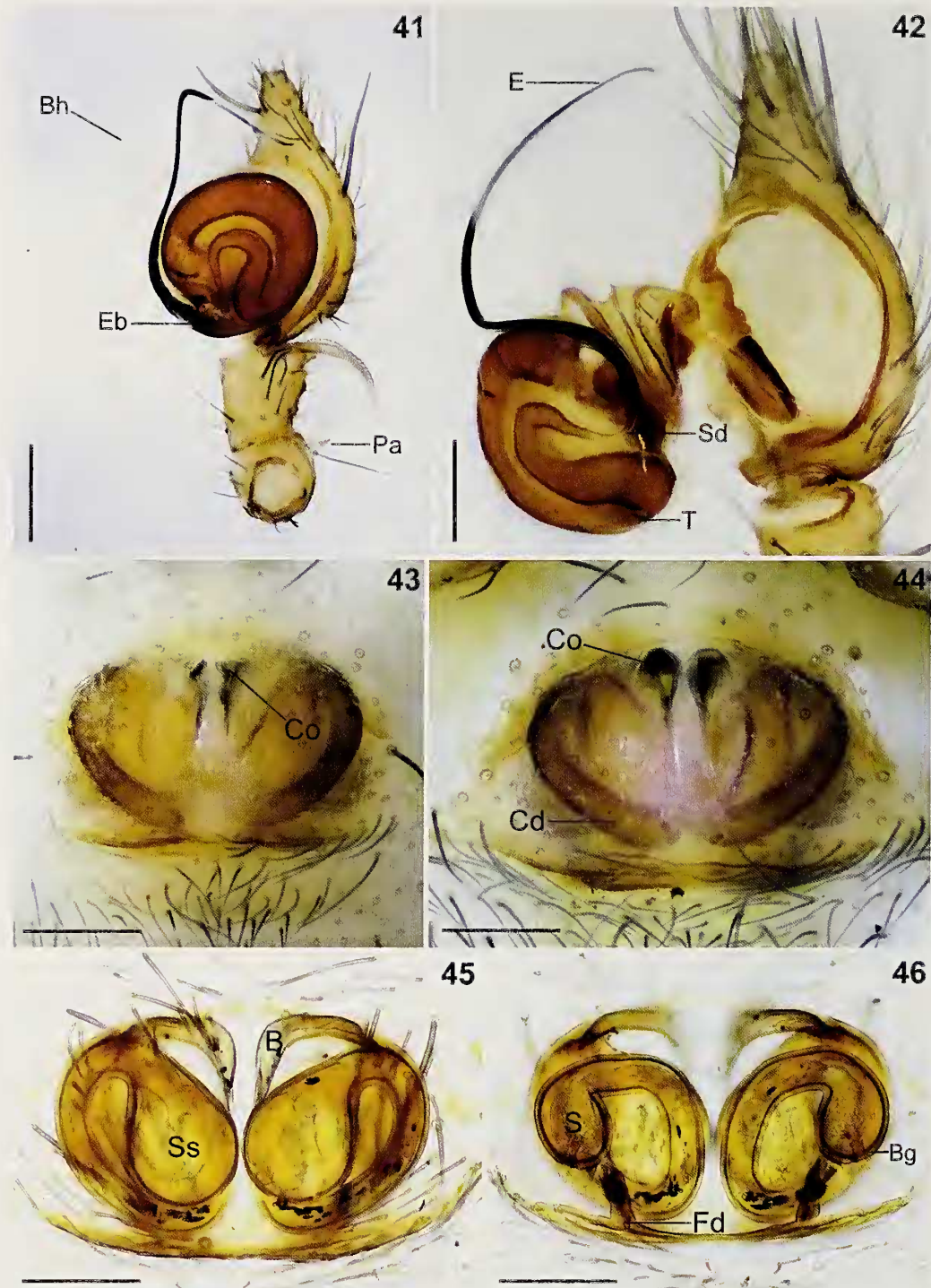
Etymology.—The specific epithet is a combination of the Latin adjective *asper* (rough) and the English noun *mbira* (African musical instrument), referring to the rough texture of the cephalothorax cuticle around the pedicel and the corresponding shape of the stridulatory organ over the anterior surface of the abdomen.

Diagnosis.—*Neoantistea aspembira* sp. nov. is similar to *N. pueblensis* Opell & Beatty, 1976, *N. inaffecta* Opell & Beatty, 1976, and *N. magna* (Keyserling, 1887) by the perpendicular

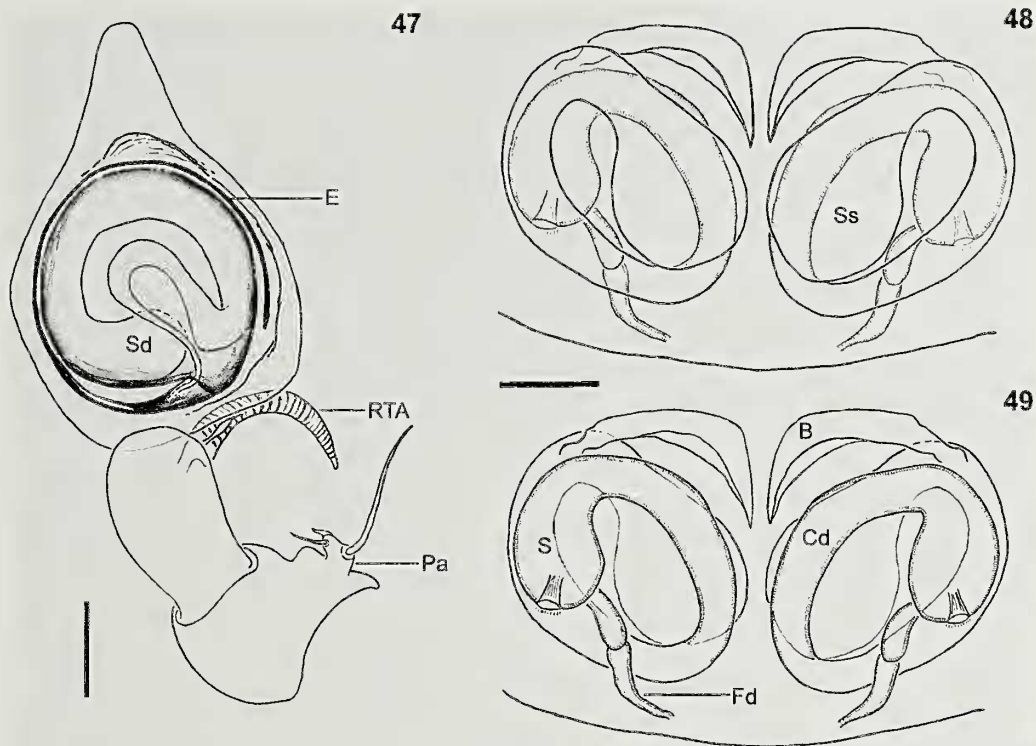
position of the RTA relative to the tibia longitudinal axis (Opell & Beatty 1976: figs. 36, 37, 68, 69, 74, 75). It differs from these species by having a distally curved RTA with serrated pattern along inner margin, and by having a short patellar apophysis which is projected dorsally with a cone-shaped tip (Figs. 47, 52, 53). The epigynum of *N. aspembira* sp. nov. has a unique anatomy characterized by an epigynum with secondary spermathecae located ventrally and larger in size than the primary spermathecae (Figs. 45, 48). Primary spermathecae oval, with almost the same width as the copulatory ducts, located laterally and posterior to the secondary spermathecae (Figs. 46, 49).

Description.—*Male* (Holotype). Total length 2.9. Cephalothorax 1.6 long, 0.95 wide. Carapace dorsal surface light brown, darker in pars cephalica; pars thoracica with radial furrows extending towards the carapace edges (Fig. 33); pear-shaped, narrowed anteriorly, fovea longitudinal and brown; inconspicuous stridulatory organ on posterior margin composed of rough area with tiny flattened hooks. Ocular region 0.24 long, 0.42 wide. Eight eyes, both eye rows slightly procurved in frontal view (Fig. 36). Eye sizes and interdistances: AME (0.07), ALE (0.12), PLE and PME equal in size (0.1); AME-AME (0.03), AME-ALE (0.01), ALE-PLE contiguous, PLE-PME (0.03), PME-PME (0.08). Clypeus greater than a diameter of AME, 0.15. Chelicerae 0.62 long, 0.30 wide, reddish; retrolateral edges slightly concave apically, three promarginal and four retromarginal teeth. Endites 0.24 long, 0.28 wide, light brown, convergent apically, slightly elongated posteriorly, serrula darker in color. Labium 0.13 long, 0.25 wide, same color pattern as the endites, with strong basal notches. Sternum 0.55 long, 0.72 wide, light brown, margin darker. Shield-shaped, truncated anteriorly, narrowed posteriorly, anterior margin undulated, lateral margins with narrow lobes opposite coxae (Fig. 35). Abdomen oval, covered with setae, raised anteriorly above posterior region of carapace, with stridulatory organ dorsolateral to the pedicel composed of two groups of wide and flat macrosetae (Figs. 34, 64, 66). Dorsal pattern greyish, with five chevron-like markings that extend towards the postero-lateral region and two semicircular spots located in anterior region (Figs. 33, 60). Ventral surface white. Colulus indistinct. Spinnerets light brown, PLS dark distally; PMS (0.2), ALS proximal segment (0.25), distal segment (0.24), PLS proximal segment (0.25), distal segment (0.24). Legs: I 3.85 (1.1) (1.3) (0.85) (0.60); II 3.81 (1.1) (1.3) (0.81) (0.60); III 3.52 (1.1) (1.1) (0.82) (0.50); IV 4.0 (1.1) (1.3) (1.0) (0.60); leg formula: 4123; light brown, precoxal triangles present, all femora and tibiae with proximal and distal dark rings. Patella I to IV dorsal surface with two macrosetae. Tibia I to IV with four macrosetae; two dorsal, one prolateral and one retrolateral. Metatarsus III–IV with four macrosetae, same distribution as tibia; tarsi without macrosetae. Pedipalp: Patella almost same length as the tibia. Retro-basal apophysis flanked by two macrosetae (Fig. 53). Tibia longer than wide. Cymbium oval, slightly longer than wide, tip slightly elongated; embolus originating pro-basally, extending around the tegulum making nearly a full turn (Fig. 38). Tegulum disk-shaped, spermatid duct following the tegular margin and with three turns before entering the embolus (Figs. 41, 47).

Female (Allotype). Total length 2.5. As in male except as noted: Cephalothorax: Carapace 1.0 long, 0.60 wide. Dorsal



Figures 41–46.—*Neoantistea aspembira* sp. nov. 41, Expanded male left pedipalp, ventral view; 42, expanded male right pedipalp, prolateral view; 43, female epigynum ventral view; 44, same ventral view with mating plug; 45, female epigynum ventral view (cleared); 46, same dorsal view. Scale bars = 0.2 mm (Figs. 41–44), 0.1 mm (Figs. 45, 46). B, bursae; Bg, Bennett's glands; Bh, basal hematodochae; Cd, copulatory duct; Co, copulatory opening; E, embolus; Eb, embolus base; Fd, fertilization duct; Pa, patellar apophysis; Sd, spermathecae; S, spermathecae; Ss, secondary spermathecae; T, tegulum.



Figures 47–49.—*Neoantistea aspembira* sp. nov., illustrations: 47, male left pedipalp, ventral view; 48, cleared female epigynum ventral view; 49, same dorsal view. Scale bars = 0.1 mm. B, bursae; Cd, copulatory duct; E, embolus; Fd, fertilization duct; Pa, patellar apophysis; RTA, retrolateral tibial apophysis; Sd, spermathecae; S, spermathecae; Ss, secondary spermathecae.

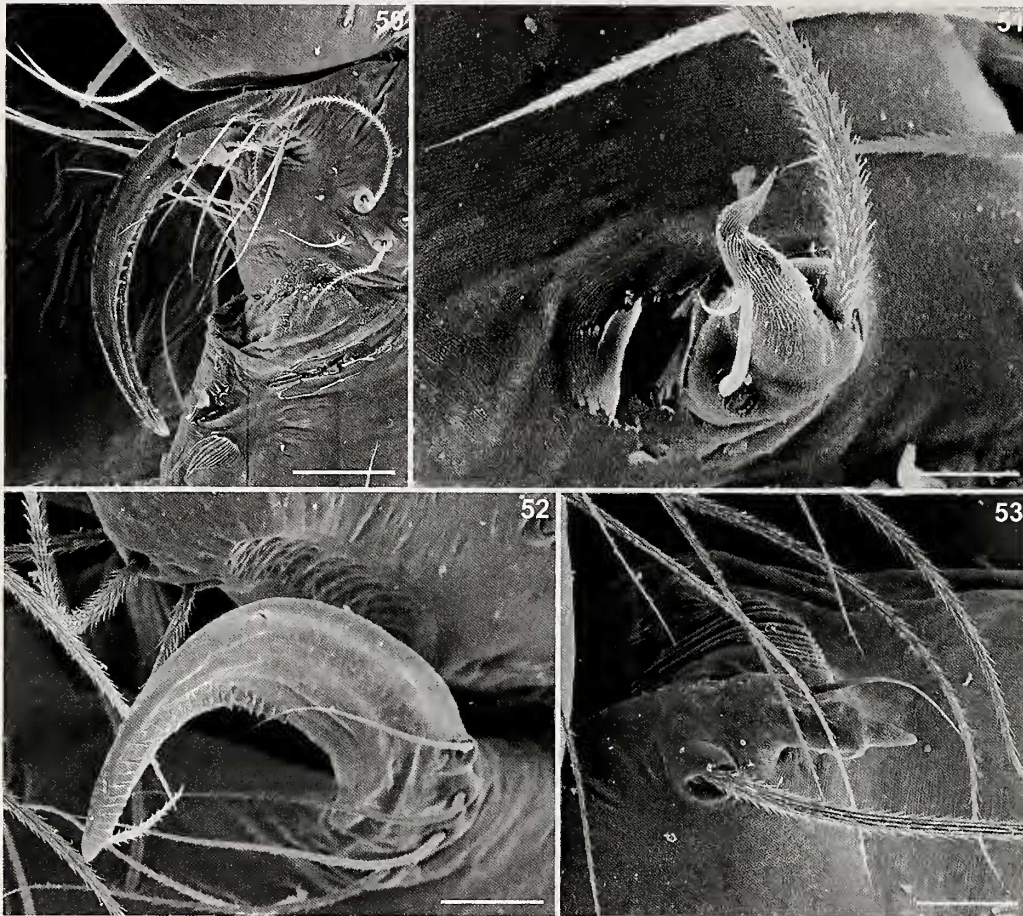
surfae light brown. Pars thoracica with a reticulated dark pattern concentrated behind the pars cephalica and over the radial furrows. Ocular region 0.15 long, 0.34 wide. Eyes and interdistances: AME (0.05), ALE (0.09), PLE (0.08), PME (0.06); PME-PME (0.07). Clypeus 0.09. Chelicerae 0.45 long, 0.20 wide; light brown. Endites 0.20 long, 0.22 wide. Labium 0.15 wide. Sternum 0.35 long, 0.52 wide. Abdomen: Tracheal system with distinctive spiracle closer to epigastric furrow than to spinnerets, internally composed of four short tubular tracheae branching in tracheoles that extend towards the posterior and anterior region of body (Fig. 54). PMS (0.15), ALS proximal segment (0.20), PLS proximal segment (0.15), distal segment (0.15). Legs: I 2.4 (0.70) (0.80) (0.50) (0.40); II 2.36 (0.70) (0.76) (0.50) (0.40); III 2.3 (0.65) (0.70) (0.55) (0.40); IV 2.77 (0.77) (0.90) (0.65) (0.45). Epigynum: flat, two copulatory openings located anterior to the spermathecae (Figs. 43, 44). Bursae separate, curved, originating dorsally from the secondary spermathecae (Figs. 45, 49). Copulatory ducts long, visible in ventral view, path partially surrounding the secondary spermathecae (Fig. 45). Fertilization ducts short and thin, originating posteriorly from the primary spermathecae (Fig. 49). The Bennett's glands are located in the posterior region of the primary spermathecae (Figs. 46, 49).

Variation ($n = 24$).—Males total length 2.1–2.9. Females total length 2.3–2.7. Coils of ducts vary slightly in orientation. Specimen coloration varies in pigmentation from whitish to reddish brown. Patterns on the dorsal surface of the carapace also vary in pigmentation from greyish to dark. Several specimens presented damaged legs.

Distribution.—Oak forest in the Atotonilco and Xamaticpac de Calchualco region, Veracruz, Mexico (Figs. 72–74).

Biology.—Male and female individuals were collected during all three expeditions. The presence of a low number of male specimens but a high number of females in October could indicate that the breeding season is during winter and spring, when more specimens were collected (23 females and 19 males in February; 31 females and 20 males in May). Juveniles are probably present in late spring to early autumn. Most specimens were caught in soil and leaf litter with pitfall traps (60 individuals), by cryptic searching (51 individuals) and with Berlese funnels (23 individuals); only four specimens were found by looking up in vegetation from the ground.

Natural history.—*Neoantistea aspembira* sp. nov. inhabits oak forest mainly in leaf litter (under the surface layer) near the bases of trees where more organic matter accumulates. Under laboratory conditions all specimens built webs over the substrate, where they spent most of their time. Structurally the webs consisted of two sheets separated by tangled middle threads (Fig. 62). The upper sheet functioned as a primary trap and was formed by unevenly spaced parallel lines crossed perpendicularly by dispersed lines that do not have any sticky property. The middle layer of threads was formed by a few lines slightly inclined connecting both sheets at one extreme of the web. The lower sheet had the same weaving as the upper sheet, but was considerably denser and had one to three openings used by the spider to either take refuge, escape or catch prey (Fig. 61). When a prey item (*Drosophila*) was caught on the upper sheet, the spider ran through one of these

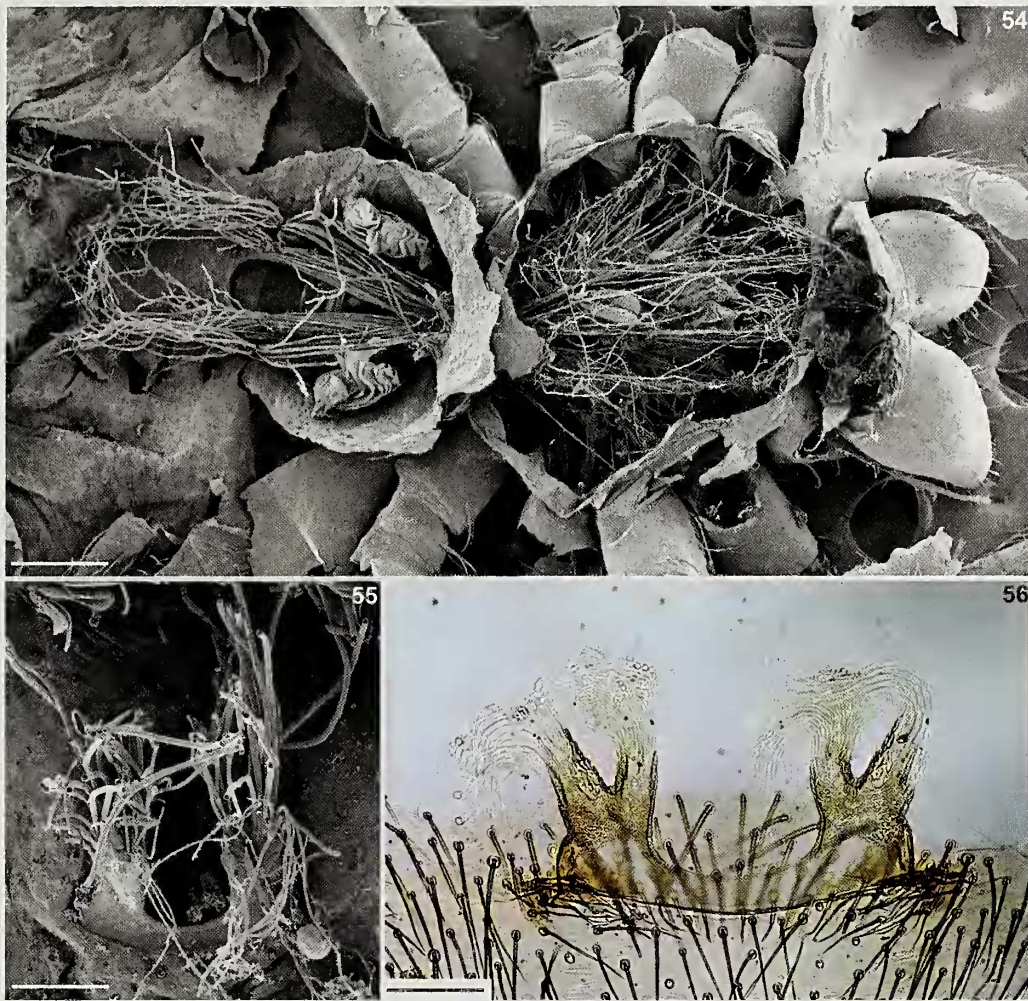


Figures 50–53.—Pedipalps of *Neoantistea* species: 50, RTA of *N. multidentata* sp. nov. dorsal view; 51, patellar apophysis of *N. multidentata* sp. nov. retrolateral view; 52, RTA of *N. aspembira* sp. nov. dorsal view; 53, patellar apophysis of *N. aspembira* retrolateral view. Scale bars = 0.01 mm.

holes, climbed the middle layer of threads and bit the fly through the upper sheet. In some cases, the lower sheet had a “shelter” formed by a small ellipsoidal web approximately the same length as the spider (2–3 mm). The total size of the web depended on the support structures and the size of the plastic cages.

Sexual behavior and reproduction.—Eight attempts at courtship between two males and seven females of *N. aspembira* sp. nov. were performed, however, only one was successful. The male courtship consisted of three basic displays: (A) chemo-exploration; (B) acoustic signaling; and (C) tactile stimulation following mating, conforming to “level two” in Foelix (2011). Chemo-exploration during the pre-courtship display (searching phase) started when the male walked around the substrate and came into contact with the female silk threads. In this phase, the male contacted the web by vertical movements of the pedipalps for approximately 10 seconds. This display is presumably used for detecting pheromones as evidenced in lycosids (Stratton & Uetz 1983; Hebets et al. 1996). During acoustic signaling, the male approached the female with vibrations and stridulations, touching the web with the pedipalps and legs in the following sequence: (1) the male produced vibrations in the web with

legs I (left) and II (right) flexed near the body; (2) leg vibrations were followed by a constant and vertical movement of the abdomen and pedicel, presumably to produce sounds with the stridulatory organ; and (3) stridulation was followed by continuous vibration of the pedipalps. These periods of activity were intercalated with periods of approximately 10 seconds of rest, after which the sequence of movements 1 to 3 were performed again, but exchanging legs I (right) and II (left) during the first phase. The male continued this behavior for approximately 35 minutes accompanied by a circular motion around the female while producing a layer of very thin threads. During tactile stimulation the male approached the female with the first pair of legs lifted, accompanied by palpal drumming (1–2 drums/sec) and stridulation by vertical and lateral movements of the abdomen. The female responded by lifting the first pair of legs and stridulating by moving the abdomen vertically while establishing contact with the male for approximately five seconds. Mating lasted approximately 48 minutes. The female remained motionless while the male, located under her abdomen, held her with the first two pairs of legs and inserted his palps: twice on the left side and twice on the right side with the left palp, and twice on the left side and twice on the right side with the right palp.



Figures 54–56.—Respiratory system of Hahniidae species: 54, Tracheae and book lungs of *N. aspembira* sp. nov., dorsal view; 55, tracheae of *H. quadriseta* sp. nov., dorsal view; 56, cleared tracheal spiracle of *N. multidentata* sp. nov., dorsal view. Scale bars = 0.1 mm (Figs. 54, 56), 0.01 mm (Fig. 55).

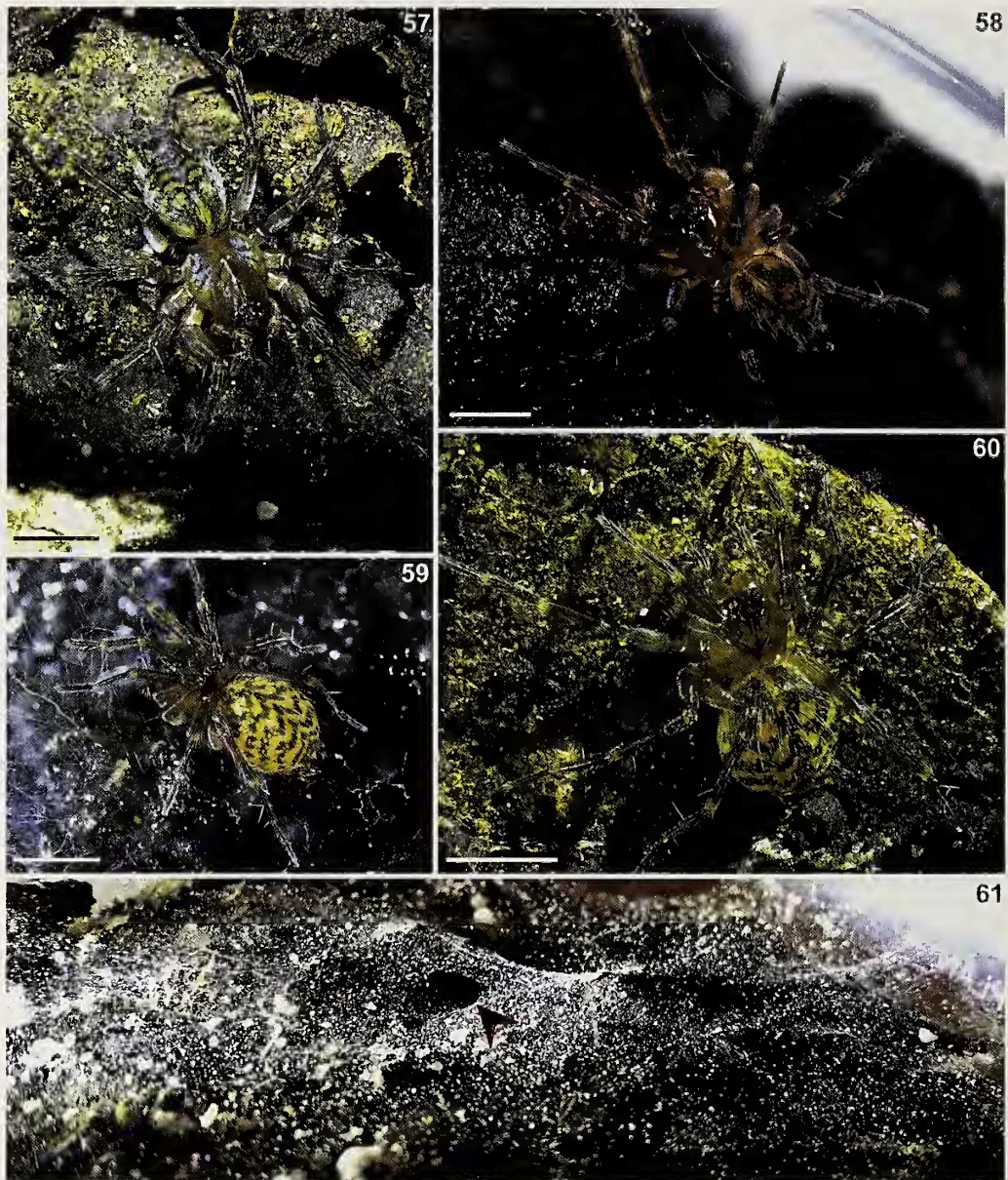
Morphology of stridulatory organs.—The new species *N. multidentata* and *N. aspembira* present simple stridulatory organs (Jocqué 2005), most likely used for intraspecific communication during species mate recognition and as defense against predators (Uhl & Elias 2011). All specimens including immatures have these structures.

The stridulatory organs are formed by antagonistic structures on the cephalothorax (*plectron*) and the abdomen (*file*) following the nomenclature of Jocqué (2005). The *plectron* structures are located on the posterior region as an elevation of the cuticle and are formed by a rough area of cuticular modifications resembling tiny flattened hooks, which are only observable with SEM microscopy (Figs. 63, 69). These hooks form discontinuous lines extending from one end to the other of this area concentrating towards the posterior edge of the carapace (Fig. 68). The *file* is located at the anterior region of the abdomen and is formed by two clusters of modified macrosetae parallel to each other and slightly oriented towards the ventral surface (Figs. 64–66); the surface of each *file* macroseta is covered by longitudinal grooves (Figs. 70, 71). The *file* clusters are separated by a space of unmodified cuticle

(ca. 0.5 x the width of the *file*) that bears two long and filiform macrosetae possibly used for receiving and decoding the vibratory signals (Figs. 65, 66).

The spatial arrangement of the *file* and *plectron* combined with the observations during courtship indicate that stridulatory sounds are produced by vertical movements of the abdomen promoting the rubbing of both structures (Fig. 67). The *N. multidentata* *file* was composed of two groups of 22 to 27 flattened macrosetae with three to four longitudinal grooves covering their total length (Fig. 70). For *N. aspembira*, the number of macrosetae is reduced to 14 to 16 per group; they are 0.3 x as long as the ones present in *N. multidentata* and provided with four to five longitudinal grooves (Fig. 71). The size differences of the *file* macrosetae between the two species suggest different frequencies of sound for species mate recognition.

The stridulatory behavior of *N. aspembira* was documented by the following five combinations: female vs. juvenile, female vs. female, male vs. female, male vs. juvenile and male vs. male. In all cases the individuals were kept in the same web, separating themselves by a distance of ca. 2 cm apart and for a



Figures 57-61.—Natural history of *N. aspembira* sp. nov.: 57, male habitus dorsal view; 58, male on the upper sheet of web; 59, female on the lower sheet of web; 60, female habitus dorsal view; 61, general aspect of *N. aspembira* sp. nov. web (lower sheet), arrow to refuge openings. Scale bars = 1 mm.

time of ca. 20 minutes. Subsequently, the individual which had been introduced moved away and built its own web. Juvenile and adult interactions ranged between one and three minutes ($2:31 \pm 0:23$) except in male-female interactions where continuous communication by the male during courtship was observed (described in the previous section). Neither aggressive behavior nor cooperation was observed among individuals; however, disputes over the same prey (*D. melanogaster*) were observed between adults vs. juveniles and females vs. males. Finally, male vs. male territorial behavior was observed, characterized by the elevation and drumming on the web by the first pair of legs.

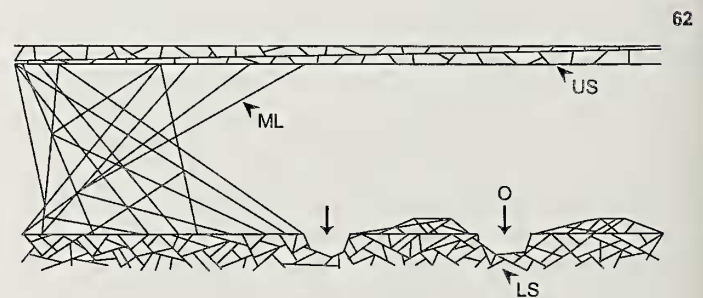


Figure 62.—Schematic representation of *N. aspembira* sp. nov. web. LS, lower sheet; ML, middle layer; O, refuge opening; US, upper sheet.



Figures 63–66.—Stridulatory organs morphology: 63, *N. multidentata* sp. nov., prosoma dorsal view, arrow to *plectron* structures; 64, *N. aspembira* sp. nov., living male individual, arrow to *file* structures; 65, *file* clusters of *N. multidentata* sp. nov.; 66, same but *N. aspembira* sp. nov. Scale bars = 0.5 mm (Fig. 63); 1 mm (Fig. 64); 0.2 mm (Figs. 65, 66).

DISCUSSION

Communication in *Neoantistea*.—The presence of stridulatory organs in both sexes and juvenile stages of *N. aspembira* suggests that communication in these spiders is not restricted to courtship and male agonistic encounters. Communication through these organs possibly plays several roles such as: avoiding other predators, during parent-offspring recognition, species recognition and social dominance interactions. Individuals of *N. aspembira* exhibited three mechanisms of acoustic signaling: stridulation, tremulation and percussion. These mechanisms have different properties that can be propagated either via airborne vibrations or through the substrate, depending on the different frequencies and amplitudes produced. Thus, the combination of different mechanisms can produce a multicomponent signal that could elicit the behaviors mentioned above (Uhl & Elias 2011).

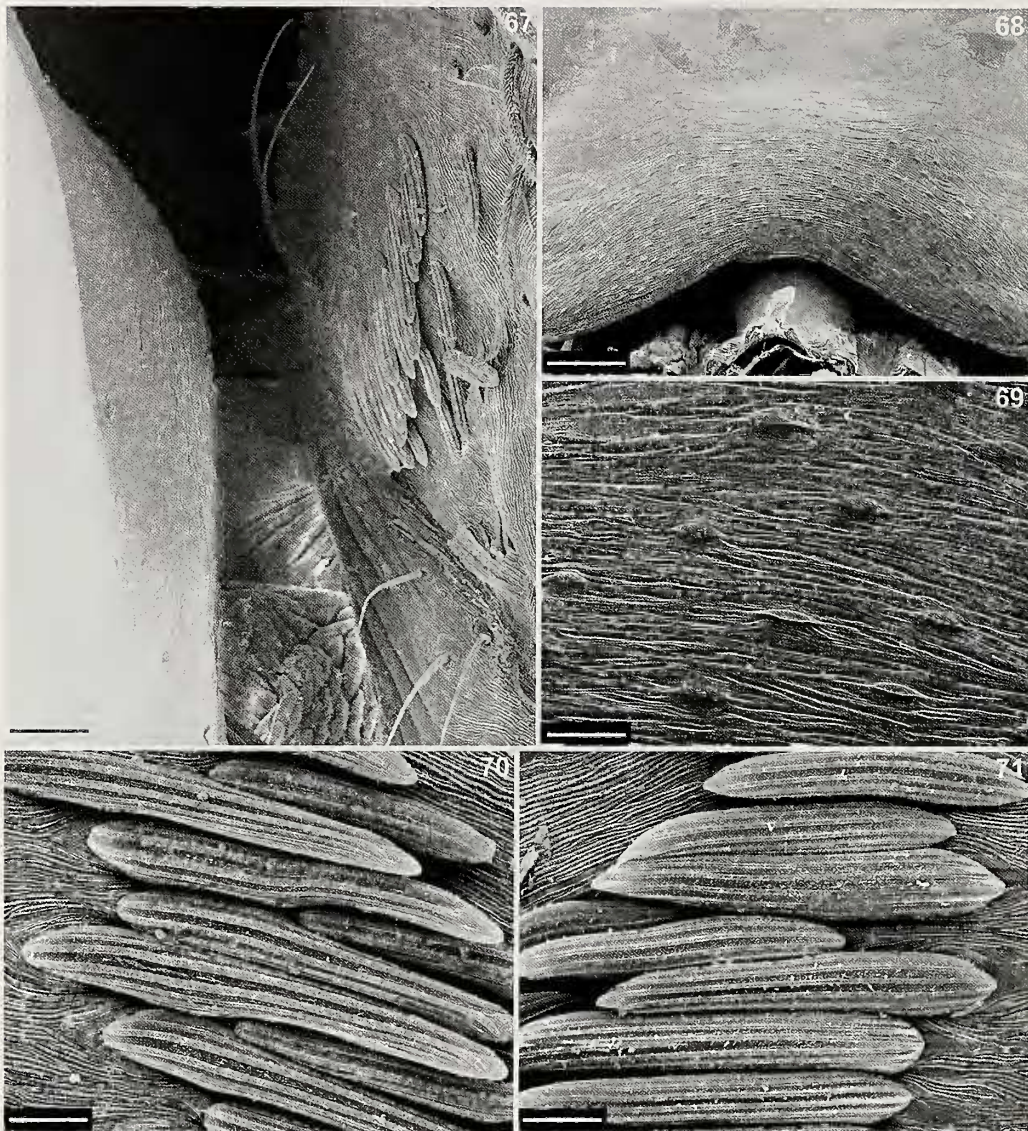
Observation of live *N. aspembira* specimens suggests that signals produced by these structures have the potential for species recognition as observed in courtship between males and females. It is important to mention that both of the newly described *Neoantistea* species are sympatric in both reproductive seasons, with more adults collected in autumn, and sharing the same microhabitat. However, differences in the size of *file* macrosetae from stridulatory organs, observation of male courtship and other somatic features differentiate them.

Importance of Mexican oak forest ecosystems.—Temperate forests in Mexico represent one of the most important

vegetation types covering approximately 24% of the country's area (CONAFOR 2009). The dominant species in Mexican temperate forest ecosystems are *Pinus* spp. and *Quercus* spp., mixed with other taxa such as: *Abies*, *Pseudotsuga*, *Picea*, *Cupressus* and *Juniperus* with a more restricted distribution (Galicia et al. 2015). Moreover, these ecosystems harbor many other species of several animal groups such as amphibians, birds, reptiles and mammals (Challenger 1998; Ceballos et al. 2002; Galieia et al. 2015).

Oak forests are mainly distributed throughout the Northern Hemisphere, and Mexico is recognized as the major diversity centre for *Quercus* with 170 native species of a total of 450 worldwide. Most Mexican oak species occur in the central and southern regions and the Sierra Madre Oriental with an estimated 109 endemic species (Rzedowski 1978; Zavala Chávez 2007; Galicia et al. 2015). The current distribution and richness of this genus in Mexico can be explained by paleoclimatic evidence and geological activity that produced numerous extinction and diversification events (Espinosa Organista et al. 2008; Galicia et al. 2015; Rodríguez Correa et al. 2015).

The diversity of Hahniidae in Mexico is represented by twelve species, most of them distributed in temperate zones, particularly in the central and southern parts of the country where up to ten species occur (Opell & Beatty 1976). The three species described here were collected as part of the first spider inventory of Mexican oak forests. This inventory and the



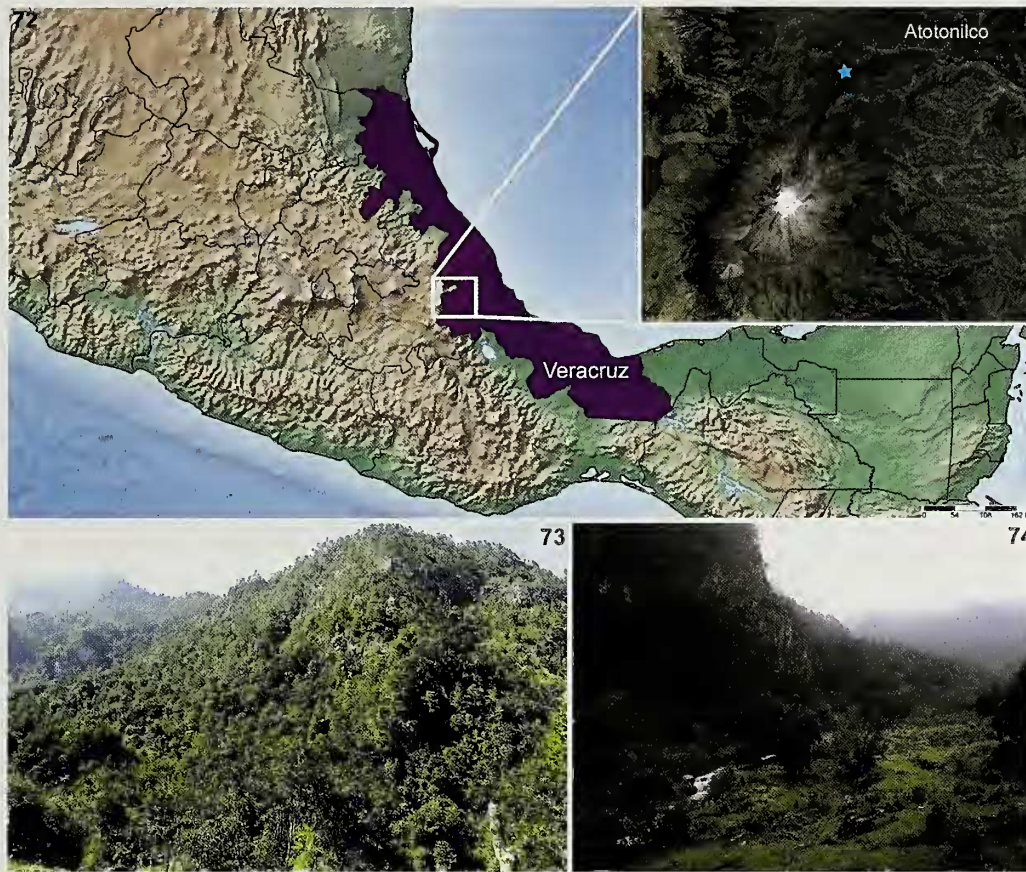
Figures 67–71.—Stridulatory organs morphology: 67, relative position of *plectron* and *file*; 68, *plectron* of *N. multidentata* sp. nov., posterior view; 69, detail of *plectron* structures; 70, *file* macrosetae of *N. multidentata* sp. nov.; 71, same but *N. aspembira* sp. nov. Scale bars = 0.01 mm.

species distributions provided by Opell & Beatty (1976) suggest that Mexican hahniids may be more diverse in oak forests, however, this conclusion is preliminary given that knowledge on hahniid diversity still is far from complete. Finally, this result predicts a potentially high spider diversity and endemism for these ecosystems, and provides data that may help to include oak forests among the protected areas of Mexico.

ACKNOWLEDGEMENTS

We would like to thank Francisco J. Salgueiro-Sepúlveda, Dulce F. Piedra-Jiménez, Mariana Servín-Pastor, Leonel Perez-Miguel, Maira S. Montejo-Cruz, Miguel Hernández-Patricio, Uriel Garcilazo-Cruz and F. Andrés Rivera-Quiroz for collecting most of the specimens during two spider inventories. Thanks to international collaborators Facundo

Labarque, Danielle Polotow and Thiago Silva for their participation in one of the 2012 expeditions. Participation by Labarque and Polotow was facilitated by Exline-Frizzell, Bill and Maria Peck, and Schlinger Arachnology fellowships from the California Academy of Sciences. Thanks also to Ali Zeltzin Lira-Olguin and Josue Lopez-Granados for helping us during the collection of live specimens, and Silvia Espinosa Matias for assisting with the SEM images. Thanks to all people from Pico de Orizaba National Park for all of their support and collaboration during expeditions, and thanks to Charles Griswold, Martín Ramírez, Cor Vink, Uriel Garcilazo-Cruz, F. Andrés Rivera-Quiroz and two anonymous referees for comments that helped to improve the manuscript. Funding was provided by UNAM-DGAPA-PAPIIT project IN213612. All species described were collected under permit SGPA/DGVS/02403/12 issued by SEMARNAT.



Figures 72–74.—Distribution map and habitat of Hahniidae species: 72, type locality; 73, *Quercus* forest at Atotonilco de Calchahualeo; 74, mountain landscape near to Pico de Orizaba Volcano. Photos: Ali Zeltzin Lira Olguin.

LITERATURE CITED

- Alvarez-Padilla, F. 2014. Cyberdiversity of Araneomorphae from Mexico. Facultad de Ciencias, UNAM. Online at <http://www.unamfearacnolab.com/>
- Alvarez-Padilla, F. & G. Hormiga. 2007 (2008). A protocol for digesting internal soft tissues and mounting spiders for scanning electron microscopy. *Journal of Arachnology* 35:538–542.
- Benoit, P.L.G. 1978. Hahniidae du Mont Kenya (Araneae). *Revue de Zoologie Africaine* 92:609–621.
- Bertkau, P. 1878. Versuch einer natürlichen Anordnung der Spinnen, nebst Bemerkungen zu einzelnen Gattungen. *Archiv für Naturgeschichte* 44:351–410.
- Bonnet, P. 1959. *Bibliographia Araneorum*. Toulouse, France.
- Bosmans, R. & E. Thijs. 1980. Spiders of the family Hahniidae from Mount Kenya. *Revue de Zoologie Africaine* 94:559–569.
- Brignoli, P.M. 1976. Ragni d'Italia XXVII. Nuovi dati su Agelenidae, Argyronetidae, Hahniidae, Oxyopidae e Pisauridae, eavernicoli ed epigei (Araneae). *Quaderni del Museo di Speleologia "V. Rivera"* 4:3–117.
- Brignoli, P.M. 1977. Sur quelques Agelenidae et Hahniidae (Araneae) d'Afrique du Nord. *Revue Arachnologique* 1:13–22.
- Brignoli, P.M. 1978. Quelques notes sur les Agelenidae, Hahniidae, Oxyopidae et Pisauridae de France et d'Espagne (Araneae). *Revue Suisse de Zoologie* 85:265–294.
- Bristowe, W.S. 1938. The classification of spiders. *Proceedings of the Zoological Society of London (B)* 108:285–322.
- Ceballos, G., J. Arroyo-Cabrales & R. Medellín. 2002. The Mammals of Mexico: composition, distribution, and conservation status. *Occasional Papers of the Museum, Texas Tech University* 218:1–27.
- Challenger, A. 1998. Utilización y Conservación de los Ecosistemas Terrestres de México. Pasado, Presente y Futuro, Comisión Nacional para el Conocimiento y Uso de la Biodiversidad, México, Mexico City.
- Chamberlin, R.V. & W. Ivic. 1942. A hundred new species of American spiders. *Bulletin of the University of Utah* 32(13):1–117.
- Chen, S.H., Y. Wang & S.C. Chen. 2003. A newly recorded spider of the family Hahniidae (Arachnida, Araneae) from Taiwan. *Journal of Taiwan normal University: Mathematics, Science & Technology* 48:25–30.
- Chen, X., H.M. Yan & C.M. Yin. 2009. Two new species of the genus *Hahnia* from China (Araneae: Hahniidae). *Acta Arachnologica Sinica* 18:66–70.
- Coddington, J.A. 1983. A temporary slide-mount allowing precise manipulation of small structures. Pp. 291–292. *In* *Taxonomy, Biology and Ecology of the Araneae*. (J.A. Coddington, ed.). *Verhandlungen des Naturwissenschaftlichen Vereins in Hamburg NP*.
- Coddington, J.A. & H.W. Levi. 1991. Systematics and evolution of spiders (Araneae). *Annual Review of Ecology and Systematics* 22:565–592.
- Coddington, J.A., C.E. Griswold, D.S. Davila, E. Penaranda & S.F. Larcher. 1991. Designing and testing sampling protocols to estimate biodiversity in tropical Ecosystems. Pp. 44–60. *In* *The Unity of Evolutionary Biology: Proceedings of the Fourth International Congress of Systematic and Evolutionary Biology*. Vol. 2. (E.C. Dudley, ed.). Dioscorides Press, Portland OR.
- Comisión Nacional Forestal (CONAFOR). 2009. *El Inventario Nacional Forestal y de Suelos de México 2004–2009*. Una

- herramienta que da certeza a la planeación, evaluación y el desarrollo forestal de México. Guadalajara, México.
- Emerton, J.H. 1890. New England spiders of the families Drassidae, Agelenidae and Dysderidae. Transactions of the Connecticut Academy of Arts and Sciences 8:166–206.
- Espinosa-Organista, D., S. Ocegueda-Cruz, C. Aguilar-Zúñiga, O. Flores-Villela & J. Llorente-Bousquets. 2008. El conocimiento biogeográfico de las especies y su regionalización natural. Pp. 33–65. *In* Capital Natural de México. Vol. I Conocimiento Actual de la Biodiversidad. (J. Sarukhan, ed.). CONABIO, Mexico City.
- Exline, H. 1938. The Arancida of Washington: Agelenidae and Hahniidae. University of Washington Publications in Biology 9:1–44.
- Foelix, R.F. 2011. Biology of Spiders. 3rd edition. Oxford University Press, New York.
- Forster, R.R. 1970. The spiders of New Zealand. Part III. Otago Museum Bulletin 3:1–184.
- Galicia, L., C. Potvin & C. Messier. 2015. Maintaining the high diversity of pine and oak species in Mexican temperate forests: a new management approach combining functional zoning and ecosystem adaptability. Canadian Journal of Forest Research 45:1358–1368.
- Gerhardt, U. & A. Kaestner. 1938. Araneae. Pp. 497–656. *In* Handbuch der Zoologie. (W. Kukenthal, & T. Krumbach (eds.). De Gruyter, Berlin.
- Gertsch, W.J. 1934. Some American spiders of the family Hahniidae. American Museum Novitates 712:1–32.
- Gertsch, W.J. 1946. Five new spiders of the genus *Neoantistea*. Journal of the New York Entomological Society 54:31–36.
- Gertsch, W.J. 1949. American Spiders. Princeton, Van Nostrand.
- Gertsch, W.J. & L.I. Davis. 1940. Report on a collection of spiders from Mexico. II. American Museum Novitates 1059:1–18.
- Harm, M. 1966. Die Deutsche Hahniidae (Arachn. Araneae). Senckenbergiana Biologica 47:345–370.
- Hebets, E.A., G.L. Stratton & G.L. Miller. 1996. Habitat and courtship behavior of the wolf spider *Schizocosa retrorsa* (Banks) (Araneae, Lycosidae). Journal of Arachnology 24:141–147.
- Jocqué, R. 2005. Six stridulating organs on one spider (Araneae, Zodariidae): is this the limit? Journal of Arachnology 33:597–603.
- Jocqué, R. & R. Bosmans. 1982. A new *Hahnia* from Ivory Coast with a note on stridulating organs in the Hahniidae (Arancida). Bulletin of the British Arachnological Society 5:319–323.
- Kaestner, A. 1968. Invertebrate Zoology, Vol. 2. Wiley Interscience, New York.
- Kaston, B.J. 1948. Spiders of Connecticut. Bulletin Connecticut Geological and Natural History Survey 70:48–54, 291–294.
- Keyserling, E. 1887. Neue Spinnen aus America. VII. Verhandlungen der Kaiserlich-Königlichen Zoologisch-Botanischen Gesellschaft in Wien 37:421–490.
- Koch, C.L. 1841. Die Arachniden. Neunter Band, Nürnberg.
- Lehtinen, P.T. 1967. Classification of the cribellate spiders and some allied families, with notes on the evolution of the suborder Araneomorpha. Annales Zoologici Fennici 4:199–468.
- Liu, N., G.Q. Huang & Z.S. Zhang. 2015. A new species of genus *Hahnia* (Araneae: Hahniidae) from South China. Zootaxa 3994:295–300.
- Marusik, Y.M. 2011. A new genus of hahniid spiders from Far East Asia (Araneae: Hahniidae). Zootaxa 2788:57–68.
- Miller, J.A., A. Carmichael, M.J. Ramírez, J.C. Spagna, C.R. Haddad, M. Řezáč, et al. 2010. Phylogeny of entelegyne spiders: affinities of the family Penestomidae (NEW RANK), generic phylogeny of Eresidae, and asymmetric rates of change in spinning organ evolution (Araneae, Araneioidea, Entelegynae). Molecular Phylogenetics and Evolution 55:786–804.
- Opell, B. D. & J.A. Beatty. 1976. The Nearctic Hahniidae (Arachnida: Araneae). Bulletin of the Museum of Comparative Zoology 147:393–433.
- Petrunkovitch, A. 1933. An inquiry into the natural classification of spiders, based on a study of their internal anatomy. Transactions of the Connecticut Academy of Arts and Sciences 31:299–389.
- Rodríguez-Correa, H., K. Oyama, I. MacGregor-Fors & A. González-Rodríguez. 2015. How are oaks distributed in the neotropics? A perspective from species turnover, areas of endemism, and climatic niches. International Journal of Plant Sciences 176:222–231.
- Rzedowski, J. 1978. Vegetación de México. Limusa, México.
- Simon, E. 1892. Histoire naturelle des araignées. Paris.
- Simon, E. 1898. Histoire naturelle des araignées. Paris 2.
- Spagna, J. & R.G. Gillespie. 2008. More data, fewer shifts: molecular insights into the evolution of the spinning apparatus in non-orb-weaving spiders. Molecular Phylogenetics and Evolution 46:347–368.
- Spagna, J.C., S.C. Crews & R.G. Gillespie. 2010. Patterns of habitat affinity and Austral/Holarctic parallelism in dieloid spiders (Araneae: Entelegynae). Invertebrate Systematics 24:238–257.
- Stratton, G.E. & G.W. Uetz. 1983. Communication via substratum-coupled stridulation and reproductive isolation in wolf spiders (Araneae: Lycosidae). Animal Behavior 31:164–172.
- Suguro, T. 2015. New data of *Hahnia* (Araneae: Hahniidae) in Japan, with a description of a new species. Acta Arachnologica 64:11–15.
- Thorell, T. 1894. Förteckning öfver arachnider från Java och närgränsande öar, insamlade af Carl Aurivillius; jemte beskrifningar å några sydasiatiske och sydamerikanska spindlar. Bihang till Kungliga Svenska Vetenskaps-Akademiens Handlingar 20:1–63.
- Uhl, G. & D.O. Elias. 2011. Communication. Pp. 127–188. *In* Spider Behaviour: Flexibility and Versatility. (M. E. Herberstein, ed.). Cambridge University Press, Cambridge.
- Wheeler, W.C., J.A. Coddington, L.M. Crowley, D. Dimitrov, P.A. Goloboff, C.E. Griswold, et al. 2016. The spider tree of life: Phylogeny of Araneae based on target-gene analyses from an extensive taxon sampling. Cladistics doi: 10.1111/cla.12182.
- World Spider Catalog. 2017. World Spider Catalog. Version 17. Natural History Museum, Bern. Online at <http://wsc.nmbe.ch/>
- Zavala-Chávez, F. 2007. Guía de los encinos de la Sierra de Tepetzotlán, México. Universidad Autónoma de Chapingo, Chapingo, México.
- Zhang, F. & C. Zhang. 2003. On two newly recorded species of the spider from China (Araneae: Salticidae, Hahniidae). Journal of Hebei University (Natural Science Edition) 23:51–54.
- Zhang, Z.S., S.Q. Li & G. Zheng. 2011. Comb-tailed spiders from Xishuangbanna, Yunnan Province, China (Araneae, Hahniidae). Zootaxa 2912:1–27.
- Zhang, Z.S. & Y.G. Zhang. 2013. Synonymy and misidentification of three *Hahnia* species (Araneae: Hahniidae) from China. Zootaxa 3682:521–533.
- Zhang, Z.S., S. Li & D.S. Pham. 2013. First description of comb-tailed spiders (Araneae: Hahniidae) from Vietnam. Zootaxa 3613:343–356.